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STUDIES TO DETERMINE WHETHER THE CONDITION OF FISH FROM

THE LOWER MACKENZIE RIVER IS RELATED TO HYDROCARBON

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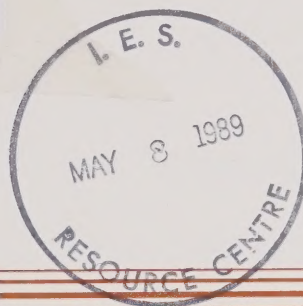
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Studies to Determine Whether the Condition of Fish from the Lower Mackenzie River is Related to Hydrocarbon Exposure

Northern Affairs Program

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1 Summary

This report was produced in response to complaints from northern people, principally residents of Fort Good Hope, that the quality of fish from the Mackenzie River (Figure 1) had deteriorated. The liver of burbot was reported to have become small and dark in colour, with the result that people would no longer eat them. The muscle of whitefish was reported to have become excessively 'watery' with the result that these fish were less palatable than in the past. Since the incidence of these complaints was coincident with operations to expand oil production at Norman Wells, this investigation was focussed on the question of whether pollution from Norman Wells (oil) could be responsible for the deterioration in the quality of the fish.

Prior to initiation of these studies, the authors had investigated the toxicological effects, both acute and chronic, of Norman Wells oil on rainbow trout in the laboratory, as part of the Freshwater Oil Spill Research Program (administered through Esso Resources, Calgary). Fish which survived exposure to the oil rejected food, grew very slowly or not at all, developed an emaciated appearance, and developed a high content of water. Previous studies with fish have reported that the water content of fish rises as the fat content falls. Preliminary examination of burbot from the Mackenzie River in 1984 showed that the small dark livers had a low fat content. Thus the laboratory trout exposed to oil developed a condition in some ways resembling that described from Mackenzie River communities, namely emaciation (low fat) and high water content.

Burbot were collected from several northern communities, from Lake Winnipeg and Southern Indian Lake, Manitoba, and from the Experimental Lakes Area in northwestern Ontario. The occurrence of small dark coloured livers was confirmed in the Mackenzie River communities, but it was not confined to communities downstream of Norman Wells. It was as severe in burbot from a small lake near Fort Franklin as it was at Fort Good Hope or Arctic Red River, and burbot would not be expected to move from Fort Good Hope to the small lake near Fort Franklin). Small, dark livers were found in the burbot from northern Manitoba also, although this condition was essentially absent from burbot from Lake Winnipeg and from the Experimental Lakes Area. If pollution is related to the liver condition, then given the distribution of the liver condition, it follows that any pollution from Norman Wells could not be the exclusive cause.

Burbot livers were analyzed for fat, protein, and moisture, and the condition was linked statistically with the fat content. In view of this, burbot from Lake Winnipeg were brought to the Freshwater Institute where an attempt was made to produce the liver condition experimentally by reducing liver fat content by simply starving the fish. Burbot were divided into two groups, one receiving food (young trout and charr) and the other receiving none. The liver fats did drop in the starved group, and livers generally became smaller and darker in colour. Thus the liver condition in burbot was produced without pollution by reducing the liver fat, although this would not rule pollution out as a cause.

Chronic laboratory exposure of trout to Norman Wells oil reduced fish growth in both length and weight, and reduced the ratio of weight to length. Furthermore, several published studies have described chronic effects of oils on growth of fish. Therefore, it seemed likely that if fish were exposed chronically to oil from Norman Wells beginning in the early 1980s, they would show body size relationships different from fish taken before that time. A collection of length/weight data on burbot caught in the early 1970s (by J.N. Stein) was compared with the current collections from the mid-1980s, and no difference was evident.

One of the most reliable indicators of a response to oil pollution in fish has been the induction of liver microsomal oxidase enzymes, and this induction was observed following experimental treatment of burbot with Norman Wells oil in the laboratory. The most diagnostic of these enzyme activities in burbot from Fort Good Hope was not notably higher than the same activity in burbot

from Lake Winnipeg. However, the presence of PCBs at higher levels in the Lake Winnipeg burbot may have compromised the liver enzyme data since some congeners of PCBs also affect these enzyme activities.

The moisture content of whitefish was less thoroughly investigated, but fish from the lower Mackenzie drainage were confirmed to have among the highest moisture values published to date for North American whitefish.

Fish from the Fort Good Hope and the other northern communities were found to contain low levels of several low-boiling hydrocarbons, notably ethylbenzene and the xylenes. These materials are among the most abundant compounds present in the 'water-soluble fraction' of Norman Wells crude oil and other crude and refined petroleum. Both burbot and whitefish taken at Fort Good Hope under winter ice-covered conditions (April, 1985) had much higher levels of these hydrocarbons than under open-water conditions (October, 1985), suggesting the likelihood of a source of these materials within the river. During the open-water season these low-boiling materials would be expected to volatilize from the water, however, during ice cover these hydrocarbons would be trapped and they would move downstream with the current.

Polycyclic aromatic hydrocarbons were also identified in some of the fish (identified only by retention time on high pressure liquid chromatography); those found most frequently were naphthalene, chrysene/benz(a)anthracene, phenanthrene, fluorene, and fluoranthene. Levels were generally similar or below those reported for burbot from the Finnish archipelago of the Baltic Sea.

Examination of tissues for organochlorine and metal contaminants indicated no unusual occurrence of metals but unexpectedly high concentrations of several organochlorines, notably toxaphene, were found.

2 Résumé

Le présent rapport a été rédigé en réponse à des plaintes formulées par les résidents du Nord, principalement ceux de Fort Good Hope, au sujet de la détérioration de la qualité des poissons du fleuve Mackenzie. On a rapporté que la population avait cessé de consommer de la lotte car le foie de ce poisson avait rapetissé et pris une teinte fncée. On a également rapporté que les muscles des corégones contenaient un surplus d'eau, ce qui rendait le poisson beaucoup moins savoureux qu'autrefois. Étant donné que le dépôt des plaintes coïncidait avec des activités visant à accroître l'exploitation pétrolière à Norman Wells, l'objet principal de l'étude a consisté à déterminer si la pollution issue de Norman Wells pouvait expliquer la détérioration de la qualité des poissons.

Avant d'entreprendre les études, les auteurs du rapport ont étudié les effets toxicologiques aigus et chronique du pétrole de Norman Wells sur des truites arc-en-ciel gardées en laboratoire, dans le cadre du Programme de recherche sur les écoulements de pétrole en eaux douces (administré par Esso Resources, Calgary). Chez les poissons qui ont survécu à l'exposition au pétrole, on a observé les réactions suivantes: rejet de la nourriture, croissance très ralentie ou inexistante, développement d'une apparence émaciée au augmentation de la teneur en eau. Des études antérieures sur les poissons avaient révélé que chez cette espèce animale, la teneur en eau augmentait lorsque la teneur en graisse diminuait. En 1984, un examen préliminaire de lottes du fleuve Mackenzie a révélé que le foie de ces poissons était petit et foncé et qu'il avait une faible teneur en graisse. Or, les truites de laboratoire ayant été exposées au pétrole avaient développé des troubles similaires à ceux décrits par les collectivités de la région du fleuve Mackenzie; soit une faible teneur en graisse (émaciation) et une forte teneur en eau.

À plusieurs endroits du Nord, on a recueilli des lottes notamment dans le lac Winnipeg, le lac sud des Indiens au Manitoba, et le secteur des lacs expérimentaux, dans le nord-ouest de l'Ontario.

3 Introduction

This investigation was undertaken jointly by the Department of Fisheries and Oceans, the Department of Indian Affairs and Northern Development, and the Dene Nation following complaints by residents of Fort Good Hope and other communities along the lower Mackenzie River (Figure 1) that the quality of fish had deteriorated recently, and that some fish were no longer suitable for domestic consumption. Liver of burbot (known locally by several names, eg. loche, ling, maria) *Lota lota*) has been consumed traditionally by people in northern communities. The quality of liver is judged by appearance and texture to be edible or not. Specifically the complaints have described a recent increase in the abundance of unusually small and dark-colored livers in burbot from several communities, and these livers are considered to be inedible. A second complaint has been that the flesh of 'whitefish' (lake whitefish, *Coregonus clupeaformis* and broad whitefish, *Coregonus nasus*) has become excessively 'watery' in consistency. Our intent was to examine whether the complaints could be described quantitatively, and whether the deterioration in the quality of the fish could be related to oil industry operations at Norman Wells.

Independently of these complaints several laboratories have been examining environmental implications of expanded oil production at Norman Wells. Environment Canada and Esso Resources have both carried out several studies. The role of our laboratory was to examine fish for the presence of several types of pollutants, and to conduct biological examinations to explain the nature and cause of the condition of the fish. We have used Norman Wells oil in studies of oil chemistry, oil toxicology, fish tainting, and enzyme induction.

Any study with a fish capable of extensive geographic movements can be confounded by unknown movements of the fish. The migratory movements of burbot in the lower Mackenzie system are not well known. Stein *et al.* [150] reported that most burbot tagged at Aklavik were recovered at the same site, but that a few (6 out of 98) moved distances of 15–49 km and one (out of 98) moved over 50 km. Movements of whitefish are known better; several species of coregonid fishes use the lower Mackenzie River as a migratory route between summer feeding areas in the Delta and along the Beaufort coastal drainages and fall spawning and overwintering areas in the Mackenzie River, its Delta, and its tributaries. These overwintering areas extend at least as far inland as Fort Good Hope for broad whitefish and as far as the Liard River for inconnu (*Stenodus leucichthys*) and Arctic cisco (*Coregonus autumnalis*).

4 Background

Our previous studies have shown that chronic exposure of young rainbow trout (*Salmo gairdneri*) to relatively high concentrations of Norman Wells crude oil resulted in greatly reduced growth rates relative to untreated fish [34]. No analyses of fats in the trout were done, but visual examination of the fish indicated an emaciated condition suggesting a low fat content. An earlier (unpublished, 1984) survey of burbot from Fort Good Hope had shown that the small dark livers were low in fat content, and so it appeared that the liver complaint might be consistent with the condition induced in the laboratory by exposure to oil. A second laboratory observation on young oil-treated trout was that they consistently became higher in water content and so it appeared that the second complaint regarding 'watery' flesh in whitefish might also be consistent with a laboratory response to oil. Oil-exposed trout also showed severe fin erosion indicating reduced resistance to bacterial attack, but this has not been reported in fish from the Mackenzie River.

The laboratory responses to oil are similar in some ways to responses resulting from starvation. A 16-week starvation experiment with large white suckers (*Catostomus commersoni*) found the water content of muscle tissue to increase and the blood lipids and blood protein to fall [80]. Love



Figure 1: Map showing sites where samples were collected

[89][pg 222-257]; [90][pg 166-229] has reviewed much published literature on starvation in fish, and while there are notable differences due to species, season, and tissue analyzed, the general responses to starvation seem to be depletion first of stored reserves of carbohydrate, then fats, and finally protein. Depletion of fat and protein is often accompanied by an increase in water content. In view of the similarities between starvation responses and the complaints about northern fish, chronic starvation experiments with burbot and whitefish were conducted.

One of the most sensitive responses to oil pollution is the induction of liver mixed-function oxygenase enzymes. Both laboratory and field studies have shown that petroleum oils contain the components required to induce fish to form increased quantities of these enzymes [122], and Norman Wells oil has induced this response in both fish [85] and marine mammals [84]. If fish in the lower Mackenzie River area were being affected by petroleum hydrocarbon pollution from Norman Wells, then this enzyme response would be expected. Unfortunately, natural petroleum inputs to the river occur in the vicinity of Norman Wells [169], and indeed, led to the discovery of oil there [16]. These natural inputs would be expected to act similarly to hydrocarbons released by industrial activity, and would make difficult the isolation of any response due to human activity from that due to natural geologic processes. However, if fish from the Mackenzie River were being exposed chronically to either natural or spilled oil, then their liver monooxygenase enzyme activities should provide a sensitive indicator of that exposure.

Norman Wells oil, like other petroleum oils, is acutely toxic to aquatic life, however, the concentrations of oil components required to kill young fish over an exposure period of 48 h were in the range of about 1 to 12 parts per million, depending on the way the oil was presented to the

fish, and on how the concentration of oil in the water was measured [82]. Given the relatively high concentrations required to kill the fish, acute mortality would not be expected in the river. The authors are unaware of any report of fish being killed by either seepage or production oils from the Norman Wells area. The fresh oil is rich in volatile components [109] and the toxicity appears to be associated with those components, notably derivatives of benzene and naphthalene. Several published studies have shown that compounds of this type are accumulated from water by aquatic organisms and also that they are readily metabolized and excreted [79, 161, 113, 174]. The striking effects of the water-soluble fraction of Norman Wells crude oil on the growth and survival of young trout [33, 34] were essentially the same as those reported by Woodward *et al.* [173] with Wyoming crude oil, and subsequently with a refined oil [174].

A common complaint following spills of petroleum products is the detection of petroleum-like tastes and odors in fishery products, reviewed recently by Motohiro [103] and by Tidmarsh *et al.* [156]. This observation was reported in whitefish following spills from the Suncor refinery into the Athabasca River in the winter of 1982, and from an overturned fuel truck into the Cameron River (NWT) in the winter of 1983 [83]. There have not been complaints of petroleum tainting of Mackenzie River fish, although these might have been expected in fish suffering from petroleum exposure. The distinctive petroleum taint has been produced in rainbow trout and arctic charr by exposing them in to Norman Wells oil in the laboratory [87].

4.1 Discussion of some possible causes

Several phenomena can be hypothesized to produce the liver condition observed in the burbot, including: (1)petroleum exposure, (2)starvation, and (3) parasitism. In appearance, the livers affected resemble those of marine cod [89][pg 224], in which '*Depletion of oil from the livers makes them appear small and red or mottled...*', which would give rise to the argument that the burbot liver problem was associated with depletion of fats. Fish which normally store fat in liver show a darkening of liver color when fat is depleted. Indeed, Love [90][pg 210] cited Rev. Samuel Ward who wrote sometime before 1776,

'The liver of ling is extremely white, so long as the fish continues in season, and abounds with a fine oil: but as soon as it goes out of season, the liver becomes very red and affords no oil'.

Indeed, it was noted very early in our examinations of burbot that livers considered poor in quality due to their dark color were low in fat content. Burbot use the liver to store fat [32, 70]; a seasonal cycle in liver fat content has been described for an inshore population of cod *Gadus morhua* from Nova Scotia, with low liver fat during winter, rising during June to peak values from from July to October, followed by a sharp drop in November with the onset of gonadal maturation [60, 61]. Seasonal or sexual cycles have not been described for burbot liver fat, but a striking seasonal cycle has been described in liver weight [21]. It seems unlikely, however, that regular cycles form the basis for the complaints since both April and October samples from Fort Good Hope contained fish with a full range of liver conditions (see Table 1). Chen [21] noted that Alaskan burbot as old as 11 years had unripe gonads in November and December, and that many of those fish also had undersized livers; indeed the range in liver indices (liver weight as a percentage of body weight) in the Alaskan burbot was about the same (1.79-12.13) as those reported in the present study.

Previous studies of the effects of petroleum on fish have generally indicated mobilization of stored energy (fat and glycogen) deposits. Sabo and Stegeman [138] reported a drop in liver lipids from 1.8% to 1.1% (39 % loss, $p < 0.001$) in *Stenotomus versicolor* following a 30-day exposure to No. 2 fuel oil. Sabo and Stegeman [138] noted reduced levels of both glycogen and neutral lipids in

micrographs of liver from *Fundulus heteroclitus* from an oiled marsh (Wild Harbour) as compared with an uncontaminated marsh (Sippewissett). There was a particularly striking reduction in the rate of incorporation of radiolabelled acetate into triglyceride by liver of the fish from Wild Harbour. Haensly *et al.* [47] examined liver of flounders from two French harbours oiled following the wreck of the Amoco Cadiz. One population (Aber Benoit) of fish showed a large drop in lipid vacuolation relative to fish from a reference site; however, the other population (Aber Wrac'h) did not. The authors pointed out that the movements of fish in the area were not well understood. Hawkes [51] fed trout food contaminated with oil and found microscopically that liver deposits of glycogen and fat became depleted. Atlantic cod (*Gadus morhua*) and winter flounder (*Pseudopleuronectes americanus*) were exposed to oil for approximately 6 months [29] with little effect on total liver lipids, although triglycerides were decreased. These authors noted that both experimental and control groups fed similarly, and so the effect on lipids was not due to changed feeding patterns. Since phospholipids and free fatty acids increased in experimental fish as triglycerides fell, the authors concluded that their results,

'strongly indicate rapid mobilization and utilization of stored pool of lipids.'

Low liver fat content could reasonably be expected to result from a chronic shortage of food, and several publications have described this for other species. Larsson and Lewander [76] described the loss of fat (triglycerides) from liver of eels (*Anguilla anguilla* L) and an accompanying but less striking reduction in the liver index during starvation periods of 95 and 145 days. Love [88] starved cod experimentally for 78 days and found that liver fat fell from about 40 percent of liver weight to about 2 percent, and turned from the normal creamy color to the red color typical of mammalian (rat) liver. Liver weight decreased and gall bladders became distended. The water content of muscle of the starved cod increased from just over 80 percent to 84-88 percent. It is noteworthy that in at least one species, brown trout (*Salmo trutta*), the changes in body composition due to diet are very sensitive to temperature; at low temperature (5.6°C) body composition was affected little by diet but at high temperature (15°C) body composition was very sensitive to diet [36]. A starvation experiment with burbot has been reported [147] but the measurements reported all related to blood composition.

In addition to starvation, some types of parasitism have also been shown to cause a reduction in liver fats. Petrushevski and Shulman [126] cited earlier Soviet studies and gave drawings of the liver of Baltic Sea cod comparing the normal condition with that resulting from infestation with the nematode *Contracoecum aduncum*. Normal livers were large and averaged 57 % fat, while heavily infested livers were small and averaged only 14.5 % fat. The condition factor (ratio of weight (g) $\times 10^5$ divided by length (mm)³) of uninfested cod was 0.941, but this fell to 0.629 for cod with 200-300 worms. Bernier [9] has found that the liver of burbot from Fort Good Hope, Arctic Red River, and Fort Simpson were infested with the nematode *Raphidascaris acus*, and the burbot from Fort Franklin with the plerocercoids of the tapeworm *Triaenophorous nodulosus*. Furthermore, the incidence of the parasites was directly related to the observed discoloration of the livers. Bykhovskaya-Pavlovskaya *et al.* [20][pg 625] cited the following Russian experience with *Raphidascaris acus* in freshwater fish,

'Heavy larval infestations of liver with larvae make (the) organ assume (a) brown color and thicken, which undoubtedly interferes with its normal function.'

Cases in which parasites of fish cause a starvation-like response in the fish are not uncommon. An additional example is that of whiting (*Merlangius merlangus*) in which infestation with the copepod *Lernaeocera branchialis* reduced the liver fats in the fish by more than 50 % [160].

Table 1: Acute toxicities of volatile hydrocarbons to aquatic animals.

Compound	96-hr LC ₅₀ to striped bass (ppm) (reference a)	48-hr LC ₅₀ to <i>Daphnia</i> <i>magna</i> (ppm) (reference b)	24-hr Toxicity thresholds to <i>Tetrahymena</i> <i>elliotti</i> (ppm) (reference c)
Ethylbenzene	4.3	2.1	—
<i>o</i> -xylene	11.	3.1	18.
<i>m</i> -xylene	9.2	9.5	56.
<i>p</i> -xylene	2.0	8.5	17.

a Benville and Korn [123]

b Bobra, Shiu and Mackay [15]

c Rogerson *et al.* [137]

Since substituted benzenes were found consistently in the fish, the biological effects of these materials was of interest. The toxicity of ethylbenzene and xylenes to fish has not been extensively studied; most studies done have been reviewed by Whipple *et al.* [168], and acutely toxic concentrations are generally in the low parts-per-million range. Some representative toxicology results with different types of aquatic animals are given in Table 1 below:

Korn *et al.* [74] investigated longer term effects of benzene in four-week exposures of striped bass (*Morone saxatilis*) and found that their high dose (mean 6 $\mu\text{L L}^{-1}$) caused a reduction in wet weight, dry weight, and body fat, while the low dose (mean 3.5 $\mu\text{L L}^{-1}$) caused only the reduction in body fat. Interestingly they noted that,

'Fish exposed to the high level attempted to feed but were unable to locate and consume their ration. Random jerking movements were observed when food was introduced. Fish exposed to the low level had some success locating the food, and approximately 50% of the pellets were consumed. Control fish successfully consumed all their ration within 5 min.'

'After 1 wk, feeding success on low- and high-dose fish started improving gradually. At the end of the study, the control and low-level fish fed normally, while the high-level fish consumed 50% of their ration.'

While oil did not interfere with feeding of cod or flounder [29] benzene did, at least temporarily, interfere with feeding of striped bass.

Based on the literature cited above, all three of the hypothetical causes (poor nutrition, parasitic infection, chronic oil intoxication) of the conditions observed in fish from the Mackenzie River and Fort Franklin can be expected to show some linkage to the energy metabolism of the fish, and it will be very difficult to argue for one cause to the exclusion of the others based on measures of energy metabolism. Since energy metabolism is so pervasive in its relation to commonly used measurements (eg growth, calorie storage, egg production etc.) these measurements may have limited

diagnostic value. Furthermore, the hypothetical causes may not be independent. Sindermann [145] has reviewed published cases in which various chemical pollutants have been suspected to have caused or enhanced various diseases in fish, and there have been instances in which oil pollution apparently resulted in outbreaks of infectious diseases in fish [101, 43]. Rainbow trout exposed to Norman Wells crude oil developed severe fin rot [33, 34]. Similar reasoning seems to apply to parasites; Esch *et al.* [37] have argued that exposure to environmental stressors can enhance the density of parasites in fish. Whipple *et al.* [168] summarized their thoughts on the effects of monocyclic aromatics with the statement,

'These petrochemicals, interacting with other pollutants, could cause quantitative reductions in the production of fish populations by decreasing growth, reproduction, egg viability, larval recruitment and survival. There are also qualitative effects on fisheries, such as condition of fish flesh and increased parasitism and disease.'

We are not aware of studies to search for a relationship between disease or parasites and MFO enzyme activities in fish. Studies on the cunner (*Tautoglabrus adspersus*) [166] have shown that starvation decreases MFO activity in fish, as it does in mammals [30]. Thus if fish in the Mackenzie River have been starving, one might have expected to see lower MFO enzyme activities than from non-starving fish. Of the various measures used to define the problems with fish, the MFO enzymes have perhaps the greatest specificity in their association with exposure to inducing chemicals.

It is inherently impossible to prove conclusively a cause and effect relationship purely from an examination of wild fish. The best that can be hoped for is an argument consistent with one or more of the hypotheses concerning possible causes of the observations in the fish, and this can help set the design for experimental studies needed to establish cause and effect relationships. The hypotheses being examined here were firstly that the condition of the fish might be consistent with chronic exposure to petroleum, and alternately that the condition of the fish might be consistent with some natural phenomena, possibly related to nutrition and/or parasitism.

5 Methods

5.1 Fish collections

Fish from four locations were collected during October 1985 by Hatfield Consultants Ltd. (201-1571 Bellevue Ave, West Vancouver, B.C.) under contract to the Dene Nation. An attempt was made to obtain 20 whitefish and 20 burbot from each of four locations, namely Arctic Red River, Fort Good Hope, Fort Simpson, and Fort Franklin, generally obtaining burbot with baited hooks and whitefish with gill nets. Upon removal from the water fish were labelled and frozen in individual plastic bags and shipped to Winnipeg for further examination by DFO staff. The intent in collecting this set of samples was to see whether the condition of the fish would be consistent with the geographic relationship of the collecting sites to a source of contamination at Norman Wells. Fort Good Hope and Arctic Red River are on the Mackenzie River downstream of Norman Wells, while Fort Simpson is well upstream of Norman Wells and Fort Franklin is off the Mackenzie River on Great Bear Lake, but still in the same watershed. Field conditions at the time of sampling precluded sampling directly in Great Bear Lake, and the Fort Franklin samples were taken from a small lake near the community. This small lake had been used historically as a float plane base and may have experienced contamination with petroleum fuels, and so its value as a 'control' may have been compromised.

Other collections of fish relevant to the study were made by DFO staff at Fort Good Hope during November, 1983; March, 1984; April, 1985; and September, 1986; at Great Slave Lake in

April, 1985; at Lake Winnipeg in February 1986; and at Fort McPherson in October, 1986. A further collection of burbot was made by staff of the Government of the Northwest Territories at Fort Simpson in April, 1986. Samples of live burbot from Lake Winnipeg were obtained from a commercial fisherman at Victoria Beach during October, 1986, and transferred to the Freshwater Institute for experimental studies.

5.2 Analyses of fish

For most purposes fish were frozen after collection and shipped to Winnipeg in coolers. When the analyses to be performed included liver microsomal enzymes, the liver was removed and fast-frozen on dry ice as soon as possible after collection, and maintained on dry ice until arrival in Winnipeg. Burbot taken in April, 1985, at Fort Good Hope were cut open in that community and several community residents were asked to judge the livers to determine whether they were acceptable for consumption. After the judgement was made the liver of each fish was frozen on dry ice. Fish from the October, 1985, collections were not used for analyses of liver enzymes since dry ice could not be shipped readily to the different communities. These fish were frozen in conventional freezers and sent frozen to Winnipeg where they were partially thawed to a semi-solid state for biological examination and for removal of samples for analyses. Each fish was assigned a laboratory number, and the species of each fish was recorded, along with measurements of fork length (mm) and weight (g). Sex and maturity were determined from examination of the internal organs. A color photograph of the internal organs of each fish was taken, and the acceptability of each burbot liver was judged by DFO staff who had been present during the examinations of fish by residents of Fort Good Hope in April of 1985, and who tried to apply comparable standards. After scoring each fish for quality, the liver of each was removed and weighed. Scales of whitefish and otoliths of burbot were taken for determination of ages.

Samples of body organs were taken for various analyses as tabulated in Table 2 although all analyses could not be done on all samples.

Table 2: Analyses performed on the tissues

Organ	Analyses
Burbot liver and whitefish muscle	Fat, moisture, protein, glycogen (1985 samples) and ash (1986 samples) Metals (Cu, Ni, Cd, Hg, Se, Zn, V) (1985 samples) Organics (hydrocarbons, organochlorines)
Burbot liver	Mixed function oxygenases (only from livers frozen on dry ice at the time of collection)

Additional portions were taken to examine for parasites and diseases, and results of that work are reported separately.

5.2.1 Quality ratings of burbot liver

Livers were examined and scored subjectively for acceptability as human food. A quantitative rating was needed for purposes of correlating appearances to biochemical measures, and so a rating system of five numerical categories was imposed arbitrarily as shown in Table 3. Of these categories, only those rated *good* (rating 5) and possibly some rated *good/marginal* (rating 4) would be acceptable for human consumption.

Table 3: Quality ratings used to describe burbot liver.

Qualitative category	Numeric value	Acceptability as human food
Good	5	Yes
Good/marginal	4	Some
Marginal	3	No
Bad/marginal	2	No
Bad	1	No

5.2.2 Moisture analysis

Tissues were oven dried to constant weight (about 72 hours) at 80°C; initial and final weights were used to calculate the percentage of weight lost, all of which was assumed to be water.

5.2.3 Ash analysis

Ash content of tissue was determined from the weight remaining after heating the tissue at 600°C for 12 hours in a muffle furnace [24].

5.2.4 Lipid (fat) analysis

Approximately 2 g of wet tissue was weighed and extracted using scaled-down version of the Bligh and Dyer procedure [14]. Tissues were extracted in and ball-milled in Teflon centrifuge tubes. For burbot liver a known volume of the methylene chloride extract containing the lipids was dried under vacuum and phosphorus pentoxide overnight, and the percent lipid was determined gravimetrically. For muscle tissue the lipid in the extract was determined spectrophotometrically at 600 nm using the reduction of Cr⁽⁺⁶⁾ to Cr⁽⁺³⁾ as described by Fales [39] with cod liver oil as the standard.

5.2.5 Nitrogen (protein) analysis

Tissues were digested by the micro Kjeldahl procedure with final ammonia measurement at 630 nm [49, 54]. Nitrogen results were converted to protein using the correction for non-protein nitrogen by the formula below, as recommended by Craig [23].

$$\text{protein} = (\text{total N} - (\text{total N} \times 0.1580)) \times 6.25,$$

5.2.6 Glycogen analysis

Fish muscle or liver (0.3 - 0.5 g) was digested by boiling in 2-3 mL of 30 % potassium hydroxide. Glycogen was precipitated by addition of 1.1 - 1.2 volumes of 95 % ethanol, then centrifuged and resuspended in 2 mL water. It was precipitated again with 2.5 mL ethanol, then dissolved in sufficient water to give a reading on the scale of the spectrophotometer. To 2 mL of the diluted glycogen solution were added 100 μ L phenol and 5 mL sulfuric acid (36 N) [31, 102]. After 30 min the colour development was read at 420 nm and the amount of glycogen was obtained by reference to a standard calibration curve prepared with authentic oyster glycogen.

5.2.7 Calculation of calorie content

Results from lipid, nitrogen, and glycogen analyses were combined to estimate the calorie content of the burbot livers. Conversion factors used were 8.49 kcal per gram of fat [24], and 5.65 kcal per gram of protein. For glycogen, the factor was 4.2 kcal per gram [72].

5.3 Analyses of mixed function oxygenase enzymes

Three mixed-function oxygenase enzyme activities were measured in microsomes prepared from those burbot livers which had been frozen on dry ice at the time of collection (Fort Good Hope, April, 1985 and September, 1986; Lake Winnipeg, February, 1986; Fort MacPherson, October, 1986). These were aryl hydrocarbon hydroxylase (AHH), ethoxyresorufin-O-deethylase (EROD), and p-nitroanisole-O-demethylase (ODM). Two additional activities, benzphetamine-N-demethylase and aminopyrine-N-demethylase were also measured in the samples from Fort Good Hope and Fort McPherson in 1986.

5.3.1 Microsomal preparation

Liver was examined for its acceptability as freshly as possible after capture and then it was removed and weighed, with care taken to avoid spillage of bile onto the liver. The liver was packed in a plastic bag and pressed between layers of dry ice to ensure rapid freezing. The frozen samples were shipped to Winnipeg in insulated coolers and stored at -60°C until analyzed. For preparation of microsomes pieces were chopped from the frozen liver and the pieces were allowed to thaw at 2-4°C until they were soft enough to be cut into smaller pieces with scissors. The small pieces were suspended with cold 0.02M HEPES(4-(2-hydroxyethyl)-1-piperazine-ethanesulfonic acid)/0.15M KCl buffer (pH 7.5) using 4 mL buffer per g of tissue in cold glass tissue homogenizers and homogenized using 5-7 passes with a motor-driven teflon pestle. The homogenate was then centrifuged at 12000 $\times$ g at 2°C for 20 min to remove insoluble matter and the supernatant was further centrifuged at 105000 $\times$ g at 2°C for 90 min to obtain a microsomal pellet. The pellet was washed and resuspended in HEPES/KCl buffer to a protein concentration between 5 and 25 mg L⁻¹. Protein in microsomal suspensions was measured by the method of Lowry *et.al.* [91] as modified by Markwell *et.al.* [97] with bovine serum albumin as a standard. Enzyme activities were measured in the resuspended microsomal preparations.

5.3.2 Aryl hydrocarbon hydroxylase (AHH)

The AHH assay was based on that of Depierre *et.al.* [28] as modified by Van Cantfort *et.al.* [159] to measure the production of polar metabolites from non-polar tritiated benz(a)pyrene used as substrate. The microsomal suspension (150 μ L) was incubated in Tris-HCl buffer (800 μ L, 55 mM, pH 7.5) with magnesium chloride (10 μ L, 500 mM) and an NADPH generating system (NADP, 10

μL , 78.7 mg mL^{-1} ; sodium isocitrate $10 \mu\text{L}$, $154.86 \text{ mg mL}^{-1}$; isocitrate dehydrogenase, q.s. 1 U mL^{-1}) and tritiated benz(a)pyrene ($20 \mu\text{L}$, $15\text{--}20 \mu\text{Ci } \mu\text{M}^{-1}$ in acetone, 1.01 mg mL^{-1}) for 30 min at 25°C . The reaction was stopped by adding 2 ml 0.15 M potassium hydroxide (KOH) in 85 % dimethyl sulfoxide (DMSO), and any benz(a)pyrene remaining unreacted was extracted with two washes, each with 3 mL hexane. Radioactivity (benz(a)pyrene metabolites) was counted in triplicate $200\text{--}\mu\text{L}$ aliquots of the remaining aqueous layer using a Beckman LS-7500 liquid scintillation counter. Metabolites were assumed to have the same specific activity as substrate and to have been produced at 1:1 stoichiometry. Three separate incubations were carried out for each microsomal suspension, and in each case triplicate blanks were run in which the microsomal suspension was boiled for 10 min prior to the addition of substrate or to which KOH-DMSO had been added prior to the addition of substrate. Radioactivity remaining in the aqueous phase in the blanks was subtracted from that in the active preparations for calculation of activity as nanomoles of product per mg of microsomal protein per minute.

5.3.3 Ethoxyresorufin-O-deethylase (EROD)

Ethoxyresorufin-O-deethylase was measured by the deethylation of 7-ethoxyresorufin to yield resorufin which was detected either by the fluorometric procedure described by Burke and Mayer [19] and Prough, Burke and Mayer [129] or that of Pohl and Fouts [128]. Using the Burke and Mayer [19] method, Tris-HCl buffer (2.75 mL , 0.1 M , pH 7.8) was mixed with magnesium chloride ($30 \mu\text{L}$, $500 \mu\text{M}$) and the NADPH generating system (NADP, $20 \mu\text{L}$, 78.7 mg mL^{-1} ; sodium isocitrate $30 \mu\text{L}$, $154.86 \text{ mg mL}^{-1}$; isocitrate dehydrogenase, q.s. 0.667 U mL^{-1}) and $220 \mu\text{L}$ of microsomal suspension was added. The reaction tube was transferred to the fluorometer (Turner 111 with primary filters 1-60 and 58 and secondary filters 23A and 10%) which was then set to zero, and $10 \mu\text{L}$ ethoxyresorufin substrate (0.07 mg mL^{-1} in dimethyl sulfoxide) was added. The fluorescence of the mixture was monitored for 1-2 min with a chart recorder, and then $10 \mu\text{L}$ of resorufin (0.125 mg mL^{-1} in dimethyl sulfoxide) was added and fluorescence was monitored for a further minute. The reaction temperature was ambient in the laboratory ($22\text{--}24^\circ\text{C}$). The activity was calculated from the rate of fluorescence development before the addition of resorufin as compared with the fluorescence of the added resorufin and expressed as nanomoles of product per mg microsomal protein per min. As before, blanks with boiled microsome preparation in the reaction mixture were also run and fluorescence of blanks (zero with this method) was subtracted from that of active preparations before calculations.

The Pohl and Fouts [128] method is similar except that a defined incubation period was used rather than continuously recording the rate of fluorescence development. The reaction mixture was 1 mL of HEPES buffer (0.1 M , pH 7.8), $10 \mu\text{L}$ of magnesium sulfate (154 mg mL^{-1}), $10 \mu\text{L}$ NADP (98.4 mg mL^{-1}), $10 \mu\text{L}$ sodium isocitrate ($193.58 \text{ mg mL}^{-1}$), isocitrate dehydrogenase (q.s. 1 U per tube), and $50 \mu\text{L}$ bovine serum albumin (40 mg mL^{-1}), all mixed in a Corex centrifuge tube and the mixture was incubated at room temperature for at least 10 min. The microsomal suspension ($50 \mu\text{L}$) was added and the reaction was initiated by the addition of $10 \mu\text{L}$ ethoxyresorufin (0.03 mg mL^{-1} , in DMSO) and the tubes were incubated for an accurately timed period of two min at 25°C . Then the reaction was stopped by the addition of 2.5 mL methanol. The samples were centrifuged at $18000 \times g$ for 5 min to remove precipitated protein and the resorufin in the supernatant was measured with an Aminco-Bowman spectrofluorometer at an excitation wavelength of 530 nm and an emission wavelength of 585 nm . Standards consisted of killed reaction mixture (methanol added) to which a known amount of resorufin had been added and which had then been carried through the centrifugation step. Blanks consisting of samples to which the methanol had been added prior to the addition of substrate were run for each sample, and the final result corrected for any non-

enzymatic production of resorufin. Triplicate incubation mixtures of each sample were run, along with triplicate blanks for each sample.

5.3.4 *p*-Nitroanisole-O-demethylase (ODM)

The analysis for O-demethylase was based on the demethylation of *p*-nitroanisole to *p*-nitrophenol and was adapted from several published methods [69, 144, 42]. Tris-HCl buffer (800 μ L; 55 mM; pH 7.5) was mixed in a Corex glass centrifuge tube with magnesium chloride (10 μ L; 500 mM) and the NADPH generating system (NADP, 10 μ L, 78.7 mg mL⁻¹; sodium isocitrate, 10 μ L, 154.86 mg mL⁻¹; isocitrate dehydrogenase q.s. 1 U mL⁻¹) and incubated at room temperature for at least 10 min. Then 50 μ L of microsomal suspension and 10 μ L of *p*-nitroanisole solution (15.3 mg mL⁻¹ in methanol) were added, and the mixture was incubated at 25°C for 20 min before being stopped by the addition of 1.5 mL of 20% trichloroacetic acid (TCA) and centrifuged at 18000 $\times$ g for 5 min. Aliquots of 0.5 mL of supernatant were transferred to clean tubes and 0.5 mL of 1M sodium carbonate was added to each and the resulting colour was read at 420 nm in a spectrophotometer *vs.* distilled water. A standard curve of nitrophenol in sodium carbonate-TCA was used to calibrate the absorbance as a function of nitrophenol concentration and blanks of boiled microsomal suspension or to which TCA had been added prior to addition of substrate were run to allow correction for any non-enzymatic hydrolysis.

5.3.5 Benzphetamine-N-demethylase (BZND)

The assay procedure for measurement of formaldehyde released was adapted from methods published previously by Prough and Zeigler [130, 92, 99, 22, 112]. The following reagents were added to Corex centrifuge tubes: 1050 μ L phosphate buffer (0.1 M, pH 7.4), 10 μ L MgSO₄ (15.4 g per 100 mL), 10 μ L NADP (98.4 mg mL⁻¹), 10 μ L sodium isocitrate (193.58 mg mL⁻¹), 10 μ L isocitrate dehydrogenase (1 Unit of ICDH activity was required. If more than 10 μ L were needed, then the volume of buffer was decreased to maintain constant total volume.), and 50 μ L semicarbazide HCl (1.338 g per 100 mL in phosphate buffer). This mixture was incubated for at least 10 minutes at room temperature, and 50 μ L of microsomal suspension were added. The sample was brought to 25°C and the reaction started by adding 10 μ L of benzphetamine hydrochloride (33.096 mg mL⁻¹ in methanol). After exactly 5 min the reaction was stopped by adding 400 μ L of zinc sulfate (15 g per 100 mL). After a further 5 min 400 μ L of saturated Ba(OH)₂ were added and the tube was centrifuged at 18000 $\times$ g for 5 min to remove precipitated protein. 1000 μ L of supernatant were transferred to a clean 12 $\times$ 75 mm test tube, and 400 μ L of Nash reagent (30 g ammonium acetate, 0.4 mL acetylacetone, and sufficient redistilled water to make 100 mL) were added. After 30 min incubation at 60°C the samples were cooled and absorbance was read against distilled water. Triplicate blanks were run on each sample by adding the ZnSO₄ and Ba(OH)₂ prior to the addition of substrate. Standards were prepared by spiking 'killed' reaction mixture with known amounts of formaldehyde.

The benzphetamine hydrochloride was a gift from the Upjohn Company, Kalamazoo, Michigan.

5.3.6 Aminopyrine-N-demethylase (APND)

This analysis was done in exactly the same fashion as benzphetamine-N-demethylase described above, except that the substrate added was 10 μ L of dimethylaminoantipyrine (8.3268 g per 100 mL in methanol).

5.3.7 Analysis for cytochrome P-450

The procedure for measuring cytochrome P-450 was based on methods described by Omura and Sato [119, 120]. Estabrooke *et.al.* [38], and Johannesen and DePierre [62] have described some of the problems with this assay. The microsome suspension was diluted to a concentration of 2 mg mL⁻¹ in the phosphate buffer (0.1 M, pH 7.4) in a 12×75 mm disposable test tube and then transferred to a quartz cuvette. The suspension was bubbled for a minimum of 20 seconds with carbon monoxide gas to saturate the sample with CO. The absorbance of the carbon monoxide-saturated microsomes was then recorded from 400 to 500 nm using a Beckman DU-7 spectrophotometer. Several milligrams of dithionite were added to the cuvette to reduce the cytochrome P-450 and the absorbance spectrum was recorded again, using the carbon monoxide spectrum as the reference. The difference absorbance readings at 450 and 490 nm were used to calculate the amount of cytochrome P-450 present, taking the difference between the absorbance at 450 and that at 490 nm, and using a millimolar extinction coefficient of 91 for a 1-cm light path [119] and the appropriate protein concentration. Analyses were done in triplicate.

5.4 Analyses of metals

For the analysis of copper, zinc, nickel, cadmium and vanadium in muscle and liver tissues, 7 to 12 g wet tissue and 10 mL concentrated nitric acid were combined in a 25 × 200 mm test tube and heated at 90°C on an aluminum hot block until dry. A further 5 mL of nitric acid was added and the drying was repeated. This addition of 5 mL of nitric acid was repeated once or twice more with liver samples high in lipid content. The final oxidation with 2 mL nitric acid and 2 mL perchloric acid proceeded until white fumes of perchloric acid were evolved, at which point all evidence of charring had disappeared. After cooling, the volume was adjusted to 25 mL with warming as necessary to redissolve all the crystals.

Copper, zinc, nickel and cadmium were analyzed by flame atomic absorption spectrophotometry with a Varian AA5 instrument with deuterium background correction. For the analysis of vanadium, cesium chloride was added to a subsample of the tissue digest to a final Cs concentration of 1 mg mL⁻¹, and vanadium was determined by direct current plasma emission spectrometry on a Spectraspan IIIB instrument with background correction.

Selenium was analyzed in 0.5 g samples of tissue as described by Vijan and Wood [163].

Mercury was analyzed in 0.2 g samples of tissue using methods of Armstrong and Uthe [5].

5.5 Extraction of low-boiling hydrocarbons

Low-boiling hydrocarbons were extracted from chopped 10-g tissues samples by passing a stream of pre-cleaned (by passage through an activated charcoal column) air through the samples maintained for 3 hr at 60°C in a water bath, as described by Murray and Lockhart [108]. After passing through the chopped fish the stream of air was then passed through a trap of activated charcoal where hydrocarbons were adsorbed. Hydrocarbons were then eluted from the charcoal with small volumes of carbon disulfide and dichloromethane which was then analyzed by capillary gas chromatography. In principle, the extraction resembled those of Berg [8] and Reinert, Hunter and Sabatino [135] except that pre-cleaned air was used as the purge gas instead of nitrogen or helium.

5.6 Chromatography of low-boiling hydrocarbons

A Varian Model 6000 gas chromatograph fitted with a flame ionization detector (FID) and a Varian Model 4270 integrator was used. The column used was a 15-m DB-5 fused silica bonded capillary

Table 4: Masses searched for with ethylbenzene and xylenes

Compound	Masses (m/e) searched for									
Ethylbenzene	106	105	103	92	91		78	77	65	51
<i>o</i> -xylene	106	105		92	91	79	78	77	65	51
<i>m</i> -xylene	107	106	105		92	91	79	78	77	51
<i>p</i> -xylene	107	106	105		92	91		78	77	51

column operated with a temperature program of 3 min at 35°C, rising to 70°C at 6°C per min, then rising to 200°C at 10°C per min, then rising to 260°C at 30°C per min, and finally holding at 260°C for 6.17 min. Nitrogen was the carrier gas at 20 cm per sec through the column. Under these conditions recoveries of ethylbenzene and xylenes added to tissues were consistently about 80 percent. Early in 1987 a second identical column, photoionization detector (PID), and data system were added to the instrument for the purpose of verifying the identification of low-boiling aromatic compounds. The ratio of responses by the two detectors has proven to be a reliable means to discriminate between aromatic and aliphatic compounds, but it has not thus far been sufficiently constant to be diagnostic for different aromatic compounds. Hence identifications are based on retention times established using authentic standards, with aromaticity confirmed by the PID.

A limited number of 1985 samples were also examined using a Hewlett-Packard gas chromatograph-mass spectrometer (model 5988) fitted with a 12-meter DB-1 capillary column. (The GC/MS instrument was owned by the University of Manitoba, Dept. of Pharmacology and was used by DFO staff on a rental basis; the column could not be changed due to the needs of other instrument users.) The identities of ethylbenzene and xylenes were based on GC retention times together with masses detected by scanning GC peaks from m/e 40 to 400 as shown in Table 4. In the cases of ethylbenzene and *o*-xylene all the mass fragments listed were present, and their pattern was clearly similar to that expected, so that the GC/MS instrument identified these compounds. In the case of *m*- and *p*-xylene (single GC peak) all the mass fragments expected were present, but early analyses also showed several additional fragments, indicating that the GC peak did not represent a single pure compound, probably the result of column bleed. Chemical ionization mass spectra using methane as a reactant gas gave M+1 peaks at m/e 107 for all three GC peaks. Later analyses with a 25-m Ultra-2 GC capillary column and a new MSD (Hewlett Packard Model 5970) installed in the Freshwater Institute gave unequivocal identifications for the xylenes. The instrument was operated in the scanning mode from m/e 40 to m/e 200 and matches to library spectra for the C₂-benzenes (ethylbenzene and xylenes) were consistently over 90 % confidence.

Samples collected before October, 1985, were analyzed by GC using a faster temperature program, and resolution of *o*-xylene from an interfering peak (unidentified then) of heptanal was inconsistent. For that reason, *o*-xylene results on samples collected before October, 1985, are excluded from the report.

5.7 Extraction of polycyclic aromatic hydrocarbons and alkanes

Five grams of liver or 15 grams of muscle were homogenized using a Waring blender and mixed with a small volume of methanol (HPLC grade). The homogenate together with 7 g KOH, 150 mL methanol, and 3 or 4 boiling chips was added to a 500-mL flask and refluxed gently for 4 hr. The cooled alkaline digest was added to a 500-mL separatory funnel with 50 mL water (HPLC grade). The mixture was shaken and extracted with three successive 50-mL portions of hexane which were combined and evaporated to 1 mL and dried with sodium sulfate. The concentrated sample was then added to the head of a 1×25 cm column packed with a slurry of 11 g activated silica gel (100-200 mesh) and 1 g alumina (5 % deactivated), with a 1- to 2-cm layer of anhydrous sodium sulfate on top. The alkane fraction was eluted from the column with 25 mL hexane, and the polycyclic aromatic fraction by 25 mL dichloromethane:hexane (1:1). The alkane fraction was evaporated and analyzed by gas chromatography, while the PAH fraction was evaporated and made up to 1 mL in acetonitrile and analyzed by HPLC.

5.8 Chromatography of polycyclic aromatic hydrocarbons

The acetonitrile solution containing the polycyclic aromatic hydrocarbons was injected into a Varian Model 5000 high pressure liquid chromatograph fitted with ultraviolet and fluorescence (Kratos FS-970) detectors. The separation of PAH was on a reverse phase Ultrasphere ODS column, 25 cm $\times$ 4.6 mm i.d., with a particle size of 5 μ m. A solvent gradient program starting with acetonitrile:water (65:35) was increased to 85:15 over 10 min and then held for 20 min. The flow rate was 1.5 mL per min, and column temperature maintained at 35°C using a heater accessory. The UV detector was set at 254 nm; the fluorescence excitation wavelength was 280 nm and emission cutoff filter was 389 nm (filter No. 389). All solvents were HPLC grade. These procedures were adapted from those described by Kinzer *et.al.* [71] (1984, EPA method 610) and Smith, Bagg and Bycroft [148].

5.9 Chromatography of alkanes

Gas chromatography conditions for separation of alkanes from C₁₂ to C₃₀ were similar to those used for low-boiling aromatic compounds described above, with the exception of the temperature program which was 2 min at 40°C followed by an increase of 10°C per min to 270°C where it was held for 6 min. Peaks were identified on the basis of retention times established by injecting authentic standards of n-alkanes. Minimum detectable concentrations were about 2 ng g⁻¹ (ppb) in tissue. With burbot liver there were numerous large unidentified peaks in the chromatograms, particularly after the retention time of eicosane (C₂₀), and these made it impossible for us to identify alkanes in that region of the chromatograms.

5.10 Analyses of organochlorines

Fish tissues were homogenized by blending with dry ice in a blender, and 10-50 g of homogenized tissue was extracted in the same manner as for PAH, except that hexane was used instead of dichloromethane (DCM). The hexane extracts were taken down to a small volume on a rotary evaporator and then taken up in hexane:DCM (1:1) for gel permeation chromatography followed by Florisil chromatography to remove biogenic lipids [116]. The Florisil column was eluted with hexane (Fraction 1) to recover PCBs, then with hexane:DCM (85:15) (Fraction 2) to recover moderately polar organochlorines, and then with hexane:DCM (1:1) (Fraction 3) to elute polar organochlorines such as heptachlor epoxide. After concentration the fractions were made to known volumes and

analyzed by gas chromatography on a Varian model 6000 instrument, equipped with an electron-capture detector and a 60-m $\times$ 0.25 mm fused silica DB-5 capillary column. The carrier gas was hydrogen at 1.6 kg cm⁻². The temperature program was from 100°C (0 min hold) to 150°C at 15°C per min then to 265°C at 3°C per min. Detector and injector temperatures were 300°C and 220°C respectively. Detector outputs were integrated with a Spectra-Physics 4270 integrator or a Varian 601 data station. PCBs were quantitated as individual isomers using standards from the National Research Council of Canada (Halifax laboratory), and chlordane, DDT, HCH and HCB were quantitated using authentic standards. One nonachlor isomer (U3) was quantitated using *t*-nonachlor. Toxaphene peaks were quantitated for the earlier samples by summing areas of six peaks in Florisil Fraction 2, identified as toxaphene-related by GC/MS, and multiplying by a response factor based on the area of the same six peaks in a toxaphene standard. Later samples were done using six peaks from Fraction 2 and two peaks from Fraction 1.

5.11 Experimental Studies

5.11.1 Induction of mixed function oxygenase enzymes

Eight large (442 mm $\pm$ 35.7 mm in length; 1213 g $\pm$ 218 g in weight) arctic charr (*Salvelinus alpinus*) were injected intraperitoneally with either corn oil (4 fish) or a mixture of corn oil and Norman Wells crude oil (1:1 by weight). The dosage was calculated from the expression:

$$\text{Volume of oil mixture, mL} = 2[(\text{body weight, kg})^{0.45}].$$

Fish were killed 4-6 days after injection and liver microsomes were analyzed for the three enzyme activities AHH, ODE, and ODM. A second similar experiment was conducted on burbot which were obtained from Lake Winnipeg in October/November, 1986, and maintained in the Freshwater Institute. Early in 1987 three burbot were injected with the same mixture of corn oil and Norman Wells crude oil as used in the experiment with charr above, and three were injected only with corn oil. Six days after injection the fish were killed and analyzed for liver MFO activities. In a third series of experiments large burbot (6 or 7 per group) were treated by intraperitoneal injection with several candidate inducers: toxaphene, 15 mg kg⁻¹ in corn oil; chlordane, 20 mg kg⁻¹ in corn oil; phenobarbital, 65 mg kg⁻¹ in normal saline; benz(a)pyrene, 30 mg kg⁻¹ in corn oil; or corn oil alone.

5.11.2 Storage stability of ethylbenzene and xylenes in tissue

Tissues were analyzed as soon as possible after arrival in the laboratory, and they were stored in freezers with temperatures ranging from -30°C to -60°C for varying periods until analysis. In order to determine the effect of storage on the analyses, 14 whitefish from Arctic Red River which had been analyzed during December, 1985, were analyzed again during July, 1986.

5.11.3 Uptake and clearance of xylene

Small arctic charr (1.365 g $\pm$ 0.303 g) were exposed to methyl-labelled ¹⁴C *o*-xylene (Pathfinder Laboratories, St. Louis) in completely filled and covered 35-L fiberglass tanks for six hr with no aeration. The concentration of *o*-xylene was measured at the beginning and end of each exposure by counting small samples of water in a liquid scintillation counter (Beckman LS-7500). There were 35 fish per tank and none died during the exposure. After 6 hr the fish were moved to clean water and groups of four fish were removed at the time of transfer and again at 23 and 45 hr after transfer. Two similar experiments were performed, one at 5°C and the other at 15°C, and fish were

acclimated to these temperatures for at least one week prior to the experiments. Radioactivity in fish was determined by burning the individual fish (in *ca.* 0.5-g portions) in a Packard sample oxidizer and counting the radioactivity in the carbon dioxide produced. Radioactivity was converted to concentration units using the specific activity of the *o*-xylene. An estimate of the half-life of *o*-xylene in the fish was made from the slope of the regression equation:

$$\ln(\text{concentration of } o\text{-xylene in fish } (\mu\text{g g}^{-1}) \text{ vs. time after transfer to clean water (hr)}).$$

5.11.4 Starvation experiment

Burbot obtained from Lake Winnipeg during October and November, 1986 were maintained in the Freshwater Institute where most fed readily on young trout and charr. Burbot were tagged early in January, 1987, and then weighed and measured individually and divided into two groups on January 28. One group continued to receive young trout or charr so that excess prey fish were always present, and the other group received no food at all. Subsamples of five fish were taken after 14, 28, 56, 112, days from both groups. The final sample from the starved group consisted of three fish only and these were frozen on days 168, 173 and 182 respectively since the fish died on those days. The final five fish from the fed group were taken on day 189 and these, together with the last three frozen from the starved group were examined on that day. The examination of each fish consisted of recording tag numbers, weighing and measuring the fish, scoring liver for quality, and taking muscle and liver samples for proximate analyses.

A parallel starvation experiment was run with lake whitefish which had been raised from eggs in the laboratory. The whitefish in the fed group were given rations of commercial fish pellets twice per week and the starved group received none. Groups of 5 fish were sampled at the onset of starvation and subsequently on after 14, 28, 56, 112, and 263 days. These fish contained extremely high quantities of visceral fat at the onset of the experiment. The starved group received an inadvertent feeding at 100 days.

5.12 Data storage and analysis

Data were stored electronically on either a VAX 11-750 or a Micro-VAX II computer using /it Datatrieve (Digital Equipment Corp.) running under VMS. Summary statistics were calculated using *MEANS* and *CORR* procedures of SAS (Statistical Analysis Services). The comparison of length/weight relationships between different groups of fish was done using the *Heterogeneity of slopes* procedure of the SAS General Linear Models procedure, as described in the SAS manual [139][pages 154-157].

6 Results and Discussion

6.1 Quality ratings of burbot liver

Inspection of burbot liver collected from Ft. Good Hope in March, 1984, confirmed the legitimacy of the complaint regarding the quality of the liver. Some livers were visibly dark in colour and small in size (Figure 2) relative to normal livers in other fish from the same catch. There is no single criterion for acceptance of liver for consumption. Livers were examined and judged subjectively to be edible or not based mainly on appearance, but some livers judged acceptable by appearance were rated as inedible because of softness in texture, small discolourations or the presence of parasites. Normally burbot liver is a pale cream colour and of uniform, firm texture, free from visible lumps or nodules. Livers showed gradations in colour and size between the extremes shown in Figure 2.

A summary of evaluations of burbot liver is given in Table 5, and it is clear that aspects of the problem with liver quality extend to several communities in the lower Mackenzie drainage, including Ft. Franklin. The problem was as prevalent at Ft. Franklin as it was at either Ft. Good Hope or Arctic Red River. It was less severe at Ft. Simpson and Great Slave Lake, and may have been qualitatively different there, and it was not found at all in burbot taken from Lake Winnipeg in February, 1986. The samples from Great Slave Lake were scored from colour photographs rather than from intact livers.

Late in 1986 several hundred burbot from Lake Winnipeg were obtained. These burbot were taken in commercial catches and kept in a holding pen for variable times until transported to Winnipeg. After arrival in Winnipeg some burbot died in the Freshwater Institute, and almost all these rated 5, but a small number were rated 3 and 4. Early in 1987 Mr. Lionel Bernier supplied a colour photograph showing a small, dark liver of a burbot from Quigley Lake in northern Manitoba. Burbot from Southern Indian Lake (Manitoba) and from the Churchill River just below Missi Falls dam in August, 1987, ranged from 2 to 4 in liver ratings. Based on these observations and on data from Alaskan burbot (see below), it seems that several northern burbot populations have some frequency of small dark livers. It appears that the changes reported in the Lower Mackenzie River basin reflects an increase in the frequency of the liver condition rather than a new phenomenon.

6.2 Burbot liver weights

Burbot livers ranged in weight from a low of 1.0 percent of total body weight to a high of 12.8 percent, a range essentially the same as that reported for burbot from the Tanana River, Alaska, in 1964 (1.79 to 12.13 percent) [21]. Chen [21] noted a seasonal cycle in burbot liver weights, with the smaller livers occurring in winter during the spawning period. Some burbot, however, had 'underdeveloped' livers and these were excluded from most of Chen's data. He suggested that burbot use liver to store nutrients in preparation for breeding.

As the winter spawning season approaches, the gonads increase in size and nutrients are withdrawn from the liver to support the growth of gonads. Adult burbot may not spawn every year [55], and Chen [21] reported taking burbot as old as 11 years from the Tanana River in Nov. and December with unripe gonads. He noted that many of these non-spawning fish had undersized livers, and suggested that the reason adult burbot fail to spawn may be because they have failed to accumulate the required nutrient supply in the liver. In view of the similarity between the liver weight indices in these burbot with those from Alaska, the comment by Chen [21][page 31] bears repeating:

'The liver weight (males and females combined) increases continuously from May to August from 4.90 to 7.85 % of body weight and then decreases all the way to 2.28 % in February. The growth of the liver can apparently be attributed to the storing of nutrients necessary for the gonad growth. As gonads begin to grow in August, nutrients are withdrawn from the liver; and the liver becomes progressively smaller and smaller'.

Most of the northern samples reported here were taken either in late winter (March/April) or late summer (September/October), and both good and bad livers were found at both times. In most groups of burbot in the present report the weight index of the liver was strongly correlated with the acceptability of the liver although this was not always the case (see below).

6.3 Proximate composition of burbot liver and muscle

Several measures of liver composition related statistically to the quality rating of the liver. The results of liver fat analyses are given in Table 6, and the ranges in liver fat levels are instructive. In

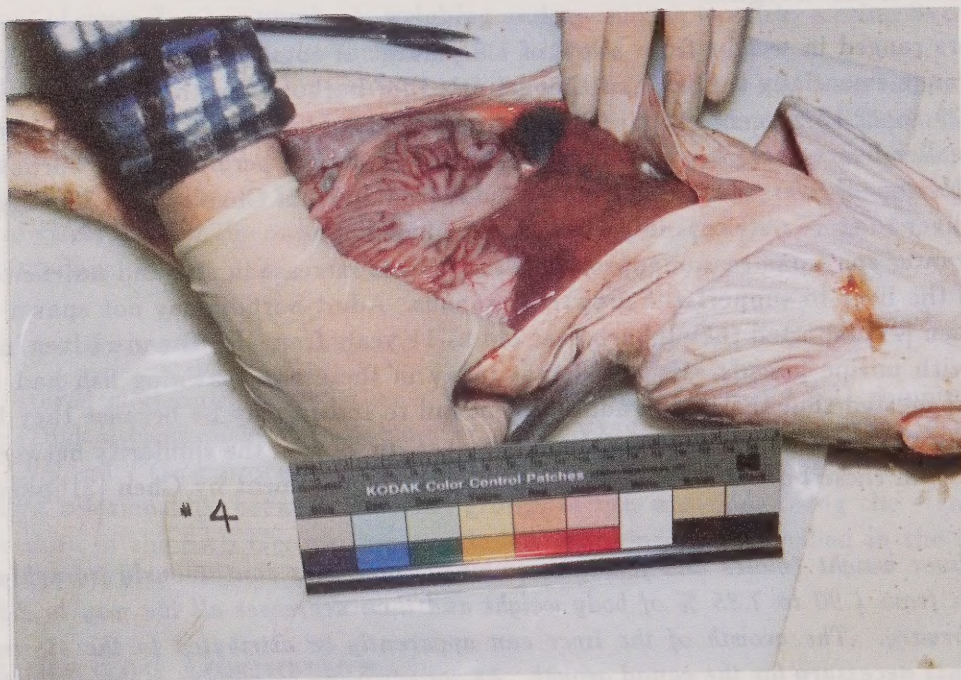
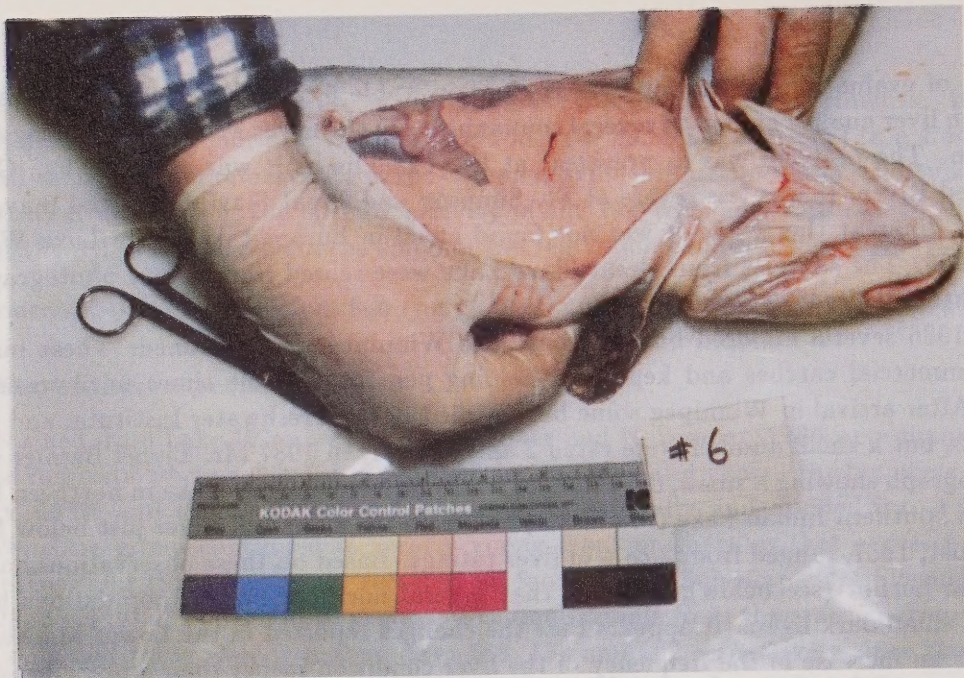


Figure 2: Representative burbot livers showing a normal liver graded acceptable for human consumption (above, No. 6), and one showing the small size and dark color graded unacceptable for consumption (below, No. 4), both collected at Fort Good Hope in April, 1985, and graded by a group of residents in that community.

Table 5: Summary of numbers of burbot livers in each quality class, based on judgement of the acceptability of liver for human consumption.

Source of fish	Date Collected	Evaluated by	Liver rating class					total	Mean score
			5 (good)	4	3	2	1 (bad)		
Ft. Good Hope	Apr '85	Community residents	4	1	0	8	3	16	2.69
Great Slave Lake	Apr '85	FWI staff (from photos)	5	4	1	0	3	13	3.62
Arctic Red River	Oct '85	FWI staff	5	0	4	1	12	22	2.32
Ft. Franklin	Oct '85	FWI staff	4	2	2	1	12	21	2.29
Ft. Good Hope	Oct '85	FWI staff	6	1	3	1	5	16	3.13
Ft. Simpson	Oct '85	FWI staff	10	3	7	0	0	20	4.15
Lake Winnipeg	Feb '86	FWI staff	25	0	0	0	0	25	5.00
Ft. Good Hope	Sep '86	FWI staff	22	6	9	6	22	65	3.00
Ft. MacPherson	Oct '86	FWI staff	3	2	1	0	0	6	4.33
Lake Winnipeg	Oct '86	FWI staff	28	2	0	0	0	30	4.87
Southern Indian Lake	Aug '87	FWI staff	0	7	2	2	0	11	3.45
Churchill River	Aug '87	FWI staff	0	2	8	3	0	13	2.92
Quality scores	Good	5							
	Good/marginal	4							
	Marginal	3							
	Bad/marginal	2							
	Bad	1							

communities reporting complaints of small, dark livers of burbot (Ft. Good Hope, Arctic Red River, Ft. Franklin) some of the fish had very low liver fat levels. In Ft. Simpson and Great Slave Lake, the extreme low fat levels were absent, although some of the livers were still rated as unacceptable, based on visible features other than small size and dark color. Correlation analyses of proximate composition data and liver acceptability data indicated that in Ft. Good Hope, Arctic Red River, and Ft. Franklin there were highly significant correlations between the acceptability ratings and the fat content of liver (Table 7); livers high in fat were much more likely to be considered edible than those low in fat. However, in Ft. Simpson and Great Slave Lake the correlations between fat and acceptability were not found. This suggests that the reason(s) for rejecting livers at Great Slave Lake and possibly at Ft. Simpson were different from those at the communities further north. It is clear that burbot liver acceptability in Ft. Good Hope, Arctic Red River, and Ft. Franklin was directly related to the fat content, with livers low in fat being low in acceptability.

Relatively few comparative data on the proximate composition of burbot have been published, especially for liver, but those available are listed in Table 8 for comparison.

There was an inverse relationship between liver fat and liver moisture in all cases, and so the negative correlations between acceptability and moisture content follow. Presumably this reflects the replacement of fat by water (or of water by fat) as described for other species [89]. Similarly, there was an inverse relationship between fat and protein, and so the negative correlation between protein and acceptability also follows (Table 7). If fat reserves were used in preference to protein (or built up only after protein had reached some required level) then we should expect protein to comprise a larger proportion of weight in those livers with low fat content.

Table 6: Proximate composition and liver calorie content of burbot.

Ft. Good Hope, March, 1984 (Lot 2) (Percent liver accounted for = 94.9)¹

Assay	Mean	SD	Range	N
Liver fat (%)	28.1	11.2	3.8 - 46.6	23
Liver protein (%)	8.68	2.41	4.58 - 14.2	23
Liver moisture (%)	58.5	10.2	40.9 - 80.4	23
Muscle moisture (%)	81.4	0.61	80.3 - 82.5	23
Liver calories (kcal g ⁻¹)	2.87	0.85	1.11 - 4.28	23

Ft. Good Hope, April, 1985 (Lot 3) (Percent liver accounted for = 90.4)¹

Assay	Mean	SD	Range	N
Liver fat (%)	27.2	15.5	3.4 - 51.8	16
Liver protein (%)	8.67	3.52	3.32 - 15.6	16
Liver moisture (%)	54.5	15.2	27.8 - 78.0	16
Liver calories (kcal g ⁻¹)	2.80	1.14	1.06 - 4.64	16
Muscle fat (%)	0.26	0.14	0.1 - 0.5	16
Muscle protein (%)	14.8	1.08	13.1 - 17.4	16
Muscle moisture (%)	82.5	2.70	78.3 - 87.8	16

Ft. Good Hope, October, 1985 (Lot 7) (Percent liver accounted for = 96.5)

Assay	Mean	SD	Range	N
Liver fat (%)	30.2	15.1	5.5 - 56.8	16
Liver protein (%)	9.12	2.36	4.42 - 14.3	16
Liver moisture (%)	56.2	12.6	33.6 - 77.5	16
Liver glycogen (mg/100g)	949.	1260.	35.6 - 3870.	16
Liver calories (kcal g ⁻¹)	3.08	1.18	1.27 - 5.07	16

Arctic Red River, October, 1985 (Lot 5) (Percent liver accounted for = 100.1)

Assay	Mean	SD	Range	N
Liver fat (%)	33.3	21.3	5.1 - 71.0	22
Liver protein (%)	9.52	3.58	3.53 - 15.5	22
Liver moisture (%)	56.6	18.4	24.7 - 80.6	22
Liver glycogen (mg/100g)	795.	1520.	41.7 - 6850.	21
Liver calories (kcal g ⁻¹)	3.36	1.63	1.19 - 6.23	22

Table 6: (continued) Proximate composition and liver calorie content of burbot.

Ft. Franklin, October, 1985 (Lot 6) (Percent liver accounted for = 98.1)

Assay	Mean	SD	Range	N
Liver fat (%)	17.4	15.1	2.5 - 49.5	21
Liver protein (%)	11.9	2.83	6.10 - 15.4	21
Liver moisture (%)	68.2	12.2	41.9 - 81.5	21
Liver glycogen (mg/100g)	597.	908.	28.4 - 2840.	21
Liver calories (kcal g ⁻¹)	2.15	1.14	0.85 - 4.64	21

Ft. Simpson, October, 1985 (Lot 8) (Percent liver accounted for = 100.3)

Assay	Mean	SD	Range	N
Liver fat (%)	37.7	7.81	27.2 - 58.2	20
Liver protein (%)	9.12	1.79	6.16 - 12.8	20
Liver moisture (%)	52.0	6.60	38.2 - 62.6	20
Liver glycogen (mg/100g)	1400.	1483.	33.7 - 5080.	20
Liver calories (kcal g ⁻¹)	3.72	0.61	2.87 - 5.34	20

Great Slave Lake, April, 1985 (Lot 4) (Percent liver accounted for = 96.5)¹

Assay	Mean	SD	Range	N
Liver fat (%)	47.0	8.91	35.2 - 63.5	13
Liver protein (%)	5.41	1.47	2.74 - 7.63	13
Liver moisture (%)	44.1	9.00	33.0 - 57.6	13
Liver calories (kcal g ⁻¹)	4.30	0.69	3.37 - 5.55	13
Muscle fat (%)	0.62	0.18	0.50 - 1.10	13
Muscle protein (%)	12.9	2.29	9.42 - 17.1	13
Muscle moisture (%)	82.1	2.02	79.3 - 86.9	13

Fort Good Hope, September, 1986 (Lot 15) (Percent liver accounted for = 85.7)¹

Assay	Mean	SD	Range	N
Liver fat	38.1	13.7	4.78 - 56.6	23
Liver protein (%)	7.78	2.55	4.57 - 15.1	23
Liver moisture (%)	38.9	15.3	10.2 - 64.6	22
Liver calories (kcal g ⁻¹)	3.68	1.03	1.26 - 5.13	23
Liver ash (%)	0.83	0.30	0.23 - 1.31	22

¹ No glycogen data

Table 7: Correlation coefficients (r) between liver acceptability ratings and liver indices (liver weight as % of body weight) or liver biochemical measurements for burbot.

Group of fish	Measurement	Mean	r	Probability
Ft. Good Hope, Apr., 1985 (Mean liver quality = 2.69) (Lot 3)	Liver index	4.62	0.59	<0.02
	Liver fat	27.2	0.62	<0.02
	Liver protein	8.67	-0.66	<0.01
	Liver moisture	54.5	-0.61	<0.02
	Liver calories	2.80	0.60	<0.02
Ft. Good Hope, Oct., 1985 (Mean liver quality = 3.13) (Lot 7)	Liver index	3.36	0.65	<0.01
	Liver fat	30.2	0.68	<0.01
	Liver protein	9.12	-0.69	<0.01
	Liver moisture	56.3	-0.60	<0.02
	Liver glycogen	949.	0.26	N S
	Liver calories	3.08	0.66	<0.01
Arctic Red River, Oct., 1985 (Mean liver quality = 2.32) (Lot 5)	Liver index	4.13	0.43	<0.05
	Liver fat	33.3	0.66	<0.001
	Liver protein	9.52	-0.49	<0.02
	Liver moisture	56.6	-0.65	<0.01
	Liver glycogen	795.	0.50	<0.05
	Liver calories	3.36	0.67	<0.001
Ft. Franklin, Oct., 1985 (Mean liver quality = 2.29) (Lot 6)	Liver index	3.98	0.84	<0.001
	Liver fat	17.4	0.78	<0.001
	Liver protein	11.9	-0.74	<0.001
	Liver moisture	68.2	-0.81	<0.001
	Liver glycogen	597.	0.54	<0.02
	Liver calories	2.15	0.77	<0.001
Ft. Simpson, Oct., 1985 (Mean liver quality = 4.15) (Lot 8)	Liver index	3.75	0.68	<0.001
	Liver fat	37.7	0.25	N S
	Liver protein	9.12	-0.66	<0.01
	Liver moisture	52.0	-0.31	N S
	Liver glycogen	400.	0.31	N S
	Liver calories	3.72	0.16	N S
Great Slave Lake, Oct., 1985 (Mean liver quality = 3.62) (Lot 4)	Liver index	6.95	0.43	N S
	Liver fat	47.0	0.26	N S
	Liver protein	5.41	-0.38	N S
	Liver moisture	44.1	-0.13	N S
	Liver calories	4.30	0.24	N S

Table 7: (Continued) Correlation coefficients (r) between liver acceptability ratings and liver indices (weight %) or liver biochemical measurements for burbot.

Group of fish	Measurement	Mean	r	Probability
Ft. Good Hope, Sept., 1986 (Mean liver quality = 3.0) (Lot 15)	Liver index			
	Liver fat	38.1	0.79	<0.001
	Liver protein	7.78	-0.71	<0.001
	Liver moisture	38.9	-0.71	<0.001
	Liver calories	3.67	0.79	<0.001
	Liver ash	0.83	-0.18	N S

Table 8: Some published values for the proximate composition of burbot.

Muscle				
Source	Fat (%)	Protein (%)	Moisture (%)	Reference
Lake Erie, (Ohio), Nov., 1955	1.2	18.0	80.6	[155]
Lake Erie, fall, 1959	0.64	16.8	80.1	[32]
Lake Huron, fall, 1961	0.6	19.2	80.2	[32]
Lake Winnipeg, June, 1948	1.05	17.5	80.8	[140]
Sand Lake, Ont., Feb., 1949	0.9	16.7	82.0	[141]
Cayuga Lake, NY, 1979	—	17.4	—	[96]
Typical value, commercial catches	1.1	17.5	81.0	[57]

Liver				
Source	Fat (%)	Protein (%)	Moisture (%)	Reference
Lake Erie, fall, 1959	58.3	6.6	34.9	[32]
Lake Winnipeg, June, 1948	35.8	—	—	[141]
Sand Lake, Ont., Feb., 1949	25.7	—	—	[142]

The range of liver glycogen values was extremely wide, from under 100 mg/100g of liver to over 7 g/100g of liver (Table 6). Significant statistical relationships existed between liver glycogen and liver acceptability in Ft. Franklin and Arctic Red River burbot, but not at Ft. Good Hope or Ft. Simpson (Table 7). Glycogen levels fall during starvation in fish, but they are also depleted rapidly during strenuous exercise [12], and so the wide range in liver glycogen content of burbot may reflect struggle during capture. It has also been shown, however, that chronic experimental exposure to petroleum in the water [100] and in the food [51] can reduce glycogen storage in fish. Fish from a contaminated harbour were depleted in glycogen relative to fish from an uncontaminated one [138].

6.4 Proximate composition of whitefish muscle

The mean water content of whitefish muscle from the October, 1985 collections ranged from 77.5 to 79.8 percent (Table 9). Only the broad whitefish from the October, 1985, sample from Ft. Good Hope were analyzed, and they fell within the range of muscle moisture shown by the lake whitefish from Ft. Good Hope in April, 1985 and those from the other communities in October, 1985. The

values are all near the upper end of the range in moisture content recorded from other Canadian and northern U.S. lake whitefish samples (Table 10). In fact, the mean value for whitefish from Arctic Red River (Table 9, 79.8 %) is the highest we have encountered. Inspection of the moisture data in Tables 9 and 10 suggests the possibility of some systematic variation in moisture content among different habitats, since ranges within our current samples of fish are relatively small, and do not overlap some of the previous measurements.

As far as the authors are aware, published data in Table 10 represented lake catches rather than river catches, and it is not certain whether that habitat difference might alter the moisture content. Two of the samples reported here (Fort Franklin, October, 1985 and Fisherman Lake, July, 1986) represent lake populations, and these had slightly lower muscle moisture levels (77.5 and 76.6 % respectively) and slightly higher muscle fat levels (2.69 and 2.05 % respectively) than the river catches. Some components of habitat are reflected in changes in body composition. For example, Hazel [52] investigated the fat content of liver in rainbow trout that had been acclimated for one month to either 5°C or 20°C, and found that liver fat was lower in the cold-acclimated fish (56 vs. 41 mg g⁻¹ liver). Perhaps the fat content of muscle of northern whitefish reflects the lower average temperatures of northern latitudes. It is apparent from Table 10 that there is a trade-off between water and fat in lake whitefish muscle ($r = -0.95$). The northern whitefish in this study have the lowest muscle fat content we have found, with the exception of the La Ronge, Sask., value from 1961 (Dugal [32], Table 10). Whitefish from the lower Mackenzie River move extensively in the river and along the coast of the Tuktoyaktuk peninsula. It is not clear how moisture content is related to quality in the domestic fishery of Mackenzie River communities, but based on the moisture content data, the complaint of 'watery' flesh of whitefish appears justified. By this reasoning the same complaint should have applied to some of the other samples in Table 10 (eg. Wapawekka L., March, 1948; Snake L., February, 1949), but the publications cited did not record any complaints.

A complaint was received in 1983 about loss of firmness in the flesh of whitefish from Great Slave Lake near the Slave River delta. In reply to the complaint Iredale (1983, personal communication) indicated that factors known to influence the textural quality of whitefish muscle included post-harvest handling, fish age, season of catch i.e. pre or immediately post spawning, and diet. Anecdotal reports have attributed soft flesh in whitefish to being left too long in gill nets so that fish caught early in the fishing period may have been dead for many hours or even days before removal from the water. It is not clear whether the complaints regarding the 'watery' condition of whitefish from the lower Mackenzie River refer to the same loss of firmness prompting the earlier complaint from Great Slave Lake, or to some additional phenomenon. The only data we have found on moisture in whitefish from Great Slave lake are those of Schmidt ([142]), and the fish had one of the lower moisture contents we are aware of (Table 10).

Love ([88]) noted a '*semi-liquid and very sloppy*' consistency in cod that had been starved experimentally for 78 days. Some pollutants also induce retention of water (edema) in fish. De Hullu and Poels [27] (cited by Helder [53]) are reported to have noted severe generalized edema in trout fry held in Rhine river water, and Helder [53] induced the condition in larval pike (*Esox lucius*) by exposing them to low levels of dioxin (2,3,7,8-tetrachlorodibenzo-*p*-dioxin). As mentioned above, the authors have observed edema in larval rainbow trout exposed to high concentrations of Norman Wells oil [33, 34].

6.5 Length of whitefish

Sampling for this study was not done with the intent of producing population growth statistics, however, since previous collections have been reported from several Mackenzie River locations

including two used in the current study (Fort Simpson and Arctic Red River), comparison with the earlier data was desirable. Stein *et al.* [150] collected fish from several sites in the Mackenzie River during 1971 and 1972, and their data on ranges for lengths of whitefish of different ages are presented in Table 11 together with lengths of fish in the 1985 collection. Lengths of most whitefish fell within ranges defined by the earlier sample. Based on this crude comparison of length ranges for the different age classes of whitefish, the striking reductions in growth typical of laboratory exposures to oil do not appear to have taken place in whitefish from Arctic Red River or Fort Simpson. Given the ages of the fish collected (7-13 years at Arctic Red River; 5-10 years at Fort Simpson) these fish would have been alive during the time of island construction and oil production expansion at Norman Wells. Growth effects have usually been observed with early life stages of fish [170] but, if these occurred they do not appear to be reflected by the current adults.

6.6 Length/Weight relationships of fish

Growth in terms of weight per unit of length was also described for several Arctic species from several locations [150], and the growth equations calculated from 1972 are given in Table 8, along with similar equations calculated from the present data. These comparisons suggest that the whitefish at Fort Simpson were considerably lighter in weight per unit of length in the early 1970s than they are now, but that otherwise differences between the two groups of fish were smaller than 10 percent. Neither whitefish nor burbot showed any striking loss in weight per unit of length relative to the earlier samples.

The length-weight relationships for several North American populations of burbot are shown in Figure 3. A more rigorous comparison of length-weight relationships was conducted using the data from 1983-86 in this report, and a set of original data from fish collected in 1970 (supplied by J. N. Stein, Department of Fisheries and Oceans, Winnipeg) (Table 13). The analysis was conducted using a SAS program to test the heterogeneity of slopes of regression lines, and it was applied to the length and weight data for burbot, lake whitefish, and broad whitefish.

The slopes of the length/weight equations were not different between the two collections for any of the species.

6.7 Starvation experiments

Subjecting burbot to prolonged starvation in the laboratory was expected to reduce the liver fat, and result in lower liver ratings, and this in fact occurred (Table 14). The liver ratings remained over 4 for the first 56 days, but fell to 2.4 by 112 days and to 1 by the end of the experiment. During the starvation period the liver weight fell from an average of over 6 percent of body weight to under 3 percent. Liver lipid content fell from about 39 percent to about 14 percent in the starved group but remained around 40 percent in the fed group. There was a close numerical relationship between the liver acceptability rating and the liver lipid content ($r=0.826$, $p<0.0001$). These results are shown graphically in Figure 4. Comparing the last two groups of fish in which the liver condition of starved fish had begun to fall below that of the fed fish (lots 23 and 25), the length/weight relationships also began to differ ($F=10.06$, $p<0.01$), whereas it did not differ earlier in the experiment.

It is clear from the starvation experiment that a condition similar in appearance to that of inedible livers can be induced in burbot without exposure to oil, and that it reflects low energy stores, principally lipids, in the liver.

With whitefish, the starvation period had no apparent effect on the composition of the muscle (Table 15). Upon dissection of these fish, they were noted to contain large reserves of adipose fat,

and these were maintained throughout the experimental period. Since these fish had been reared from eggs in the laboratory they may not provide a good model for the effects of starvation on wild whitefish.

Table 9: Proximate composition of whitefish.

	Mean	SD	Range	N
Muscle fat (%)	4.01	3.28	1.10 - 10.7	12
Muscle protein (%)	16.5	2.69	10.35 - 20.5	23
Muscle moisture (%)	78.4	1.88	75.2 - 82.7	25
Muscle glycogen (mg/100g)	28.4	52.7	4.79 - 272.	25

Lake whitefish, Ft. Good Hope, April, 1985 (Lot 3)

	Mean	SD	Range	N
Muscle fat (%)	1.61	0.78	0.7 - 2.90	7
Muscle protein (%)	14.9	0.77	14.2 - 16.5	7
Muscle moisture (%)	79.0	1.52	76.8 - 81.4	7

Lake whitefish, Arctic Red River, October, 1985 (Lot 5)

	Mean	SD	Range	N
Muscle fat (%)	1.86	0.83	0.70 - 3.60	16
Muscle protein (%)	14.5	1.50	12.0 - 17.8	16
Muscle glycogen (mg/100g)	8.61	4.33	4.72 - 21.9	16
Muscle moisture (%)	79.8	1.36	76.9 - 81.8	16

Lake whitefish, Ft. Franklin, October, 1985 (Lot 6)

	Mean	SD	Range	N
Muscle fat (%)	2.69	1.92	0.80 - 6.90	19
Muscle protein (%)	17.2	2.42	12.9 - 22.1	19
Muscle glycogen (mg/100g)	27.7	73.9	4.85 - 332.	19
Muscle moisture (%)	77.5	1.92	74.5 - 83.0	19

Lake whitefish, Ft. Simpson, October, 1985 (Lot 8)

	Mean	SD	Range	N
Muscle fat (%)	1.92	0.98	0.8 - 4.8	19
Muscle protein (%)	18.1	2.14	15.1 - 22.7	19
Muscle glycogen (mg/100g)	8.30	2.79	4.41 - 15.7	19
Muscle moisture (%)	77.6	2.00	74.4 - 81.6	19

Table 9: (Continued) Proximate composition of whitefish.

Lake whitefish, Fisherman Lake, N.W.T., July, 1986 (Lot 26)

	Mean	SD	Range	N
Muscle fat (%)	2.05	1.11	0.77 - 4.1	8
Muscle protein (%)	17.4	3.32	10.4 - 20.2	8
Muscle moisture (%)	76.6	0.79	74.9 - 77.6	8
Muscle ash (%)	1.44	0.25	1.21 - 1.9	8

Broad whitefish, Ft. Good Hope, September, 1986 (Lot 15)

	Mean	SD	Range	N
Muscle fat (%)	1.24	0.65	0.46 - 2.6	10
Muscle protein (%)	13.6	2.18	10.5 - 17.4	11
Muscle moisture (%)	79.2	1.87	76.5 - 82.8	11
Muscle ash (%)	1.28	0.10	1.11 - 1.4	11

Table 10: Some published values for the proximate composition of whitefish muscle.

Description	Fat (%)	Protein (%)	moisture (%)	Reference
Lake Huron, spring 1959	11.7	18.4	70.3	[32]
Lake Huron, spring 1961	16.3	17.4	65.6	[32]
Lake Superior, 1961	3.4	18.9	77.0	[32]
Cedar Lake (Man.), winter, 1962	3.9	18.5	76.8	[32]
La Ronge (Sask.), winter, 1961	1.7	—	—	[32]
Snake Lake (Sask.), Feb., 1949	3.05	16.9	79.2	[142]
Great Slave Lake, Jan., 1950	13.6	15.5	71.3	[142]
Wapawekka L. (Sask.), Mar., 1948	2.3	17.2	79.0	[140]
Lake Winnipeg, June, 1948	9.6	17.2	72.6	[141]
Lake Erie, (Ohio), Nov., 1951	9.5	16.3	71.5	[155]
Red Lake, (Minn.), Aug., 1954	5.4	18.3	75.1	[155]
Red Lake, (Minn.), Aug., 1954	5.9	18.4	73.7	[155]
Typical value, commercial catches	10.7	19.1	69.2	[57]

Table 11: Length ranges for fish of different age classes in 1972¹ and in 1985

Lake Whitefish Arctic Red R	1972		1985	
	Length (mm)	N	Length (mm)	N
Age 7	285 - 388	13	310 - 367	3
Age 8	316 - 370	6	378 - 400	2
Age 9	331 - 442	11	368 - 430	4
Age 10	328 - 420	5	362 - 447	3
Age 11	390 - 556	10	459	1
Age 12	421 - 477	4	420	1
Age 13	402 - 461	4	458 - 463	2

Lake Whitefish Ft. Simpson	1972		1985	
	Length (mm)	N	Length (mm)	N
Age 5	266 - 349	11	280	1
Age 6	255 - 417	15	310 - 360	2
Age 7	303 - 466	18	322 - 377	7
Age 8	352 - 460	10	329 - 373	6
Age 9	388 - 484	6	370 - 404	2
Age 10	363 - 518	7	406	1

¹From Stein *et al.* [150]

Table 12: Length-weight relationships for combined sexes of fish from Arctic Red River and Fort Simpson, calculated from fish captured in 1972¹ and from those captured in Oct, 1985, for this study.

Equations are of the form:

$$\ln(\text{weight, g}) = \text{slope}(\ln \text{length, mm}) + \text{intercept}$$

Species and date	Source and number	slope	intercept	Calculated weight of 500-mm fish
Lake whitefish, 1972	Arctic Red R., n=370	3.375	-13.402	1943
Lake whitefish, 1985	Arctic Red R., n=16	3.621	-14.909	1986
Lake whitefish, 1972	Ft. Simpson, n=50	4.277	-19.295	1458
Lake whitefish, 1985	Ft. Simpson, n=19	3.490	-14.091	1994
Burbot, 1972	Arctic Red R., n=49	3.082	-12.488	785
Burbot, 1985	Arctic red R., n=22	2.958	-11.818	710
Burbot, 1972	Ft. Simpson, n=10	2.468	-8.703	761
Burbot, 1985	Ft. Simpson, n=20	3.173	-13.055	784

¹From Stein *et al.* [150]

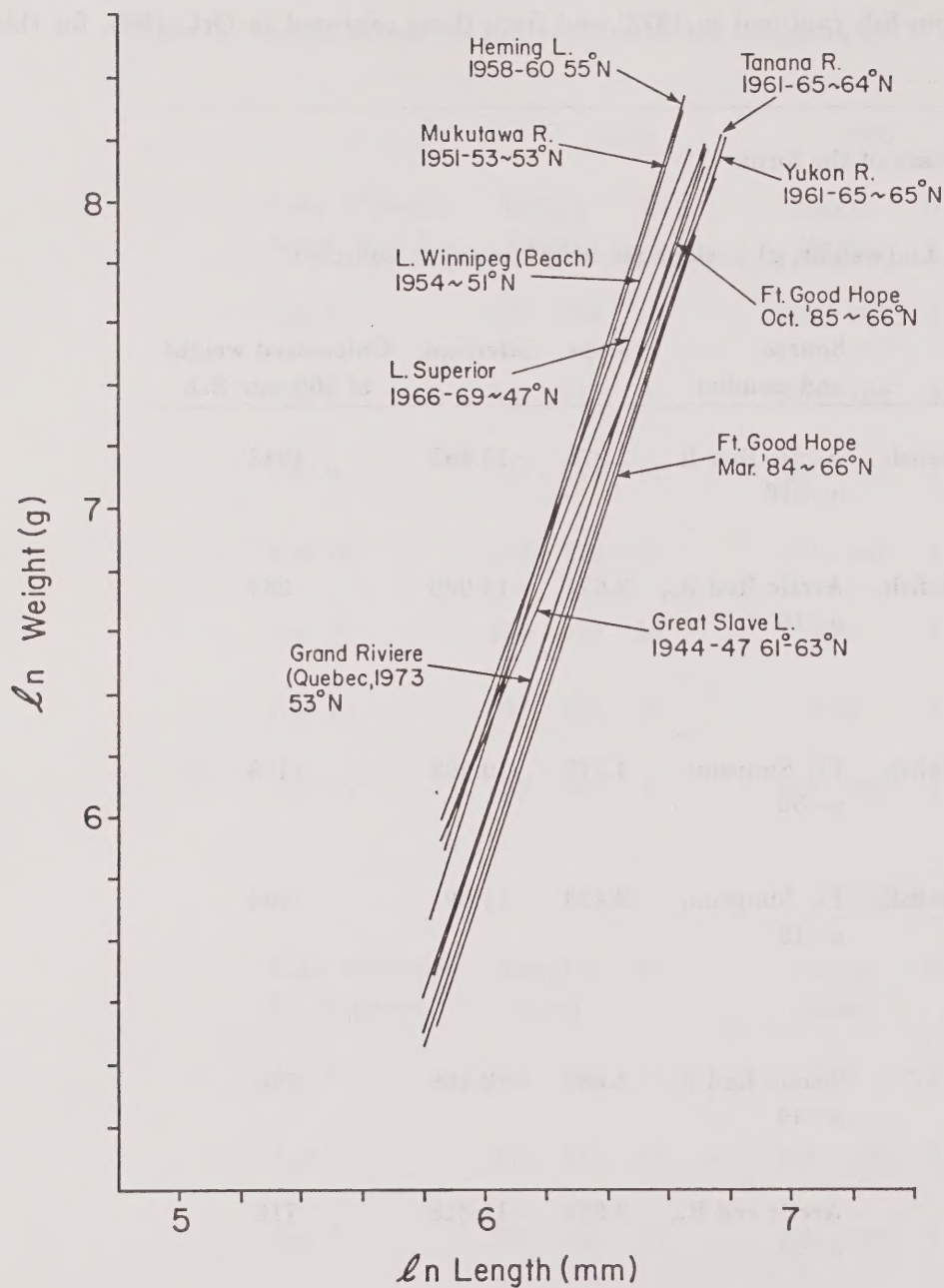


Figure 3: Logarithmic length-weight relationships for several collections of North American burbot, calculated from published data, as compared with data on burbot from the collections reported here.

Table 13: Comparison of slopes of length-weight equations for burbot and whitefish from Arctic Red River and Fort Simpson in 1970¹ and in 1983-86.

Species	Location	No. fish 1970	Slope 1970	No. fish 1983-86	Slope 1983-86	F
Burbot	A.R.R.	36	2.916	157	3.084	1.55 ²
Burbot	Ft. Sim.	10	3.132	28	3.112	0.01
Lake whitefish	A.R.R.	458	3.308	52	3.244	0.09
Lake whitefish	Ft. Sim.	470	3.182	19	3.490	0.22
Broad Whitefish	A.R.R.	585	3.181	46	3.361	0.19

¹From Stein *et al.* [150]

²Not significant

Table 14: Proximate composition and quality ratings of liver of burbot from laboratory starvation experiment, as compared with burbot maintained under similar conditions and fed young trout and charr.

Fed (control) burbot, liver						
Day	Lot	Moisture %	Protein %	Lipid %	Ash %	Liver quality
Day 0	18	42.3	5.36	38.7	0.73	4.40
Day 14	19	58.1	7.32	28.8	0.91	3.00
Day 28	20	40.6	5.86	41.5	0.73	4.40
Day 56	21	34.0	4.47	35.9	0.59	4.60
Day 112	23	38.9	4.59	44.9	0.49	4.00
Day 189	25	36.2	3.68	43.8	0.66	4.20

Starved burbot, liver						
Day	Lot	Moisture %	Protein %	Lipid %	Ash %	Liver quality
Day 0	18	42.3	5.36	38.7	0.73	4.40
Day 14	19	46.4	6.90	36.2	0.72	4.00
Day 28	20	42.2	6.35	37.0	0.82	4.20
Day 56	21	38.2	6.42	39.5	0.71	4.20
Day 112	23	56.1	9.88	20.0	0.52	2.40
Day 189	25	61.0	9.78	13.8	-	1.00

Table 15: Proximate composition of whitefish muscle from the laboratory starvation experiment, as compared with whitefish maintained under similar conditions and fed regularly on commercial fish food.

		Fed (Control) whitefish, muscle					
Day	Lot	Moisture %	Protein %	Lipid %	Ash %	Calories (kcal g ⁻¹)	Total %
Day 0	18	78.3	15.6	1.82	1.29	1.04	97.1
Day 14	19	74.0	19.2	2.91	1.61	1.33	97.7
Day 28	20	77.8	15.9	1.54	1.29	1.03	96.5
Day 56	21	75.5	16.0	5.50	1.58	1.37	98.6
Day 112	23	73.1	16.0	3.29	1.72	1.19	94.1
Day 263	31	74.4	14.3	1.71	1.49	0.95	91.8

		Starved whitefish, muscle					
Day	Lot	Moisture %	Protein %	Lipid %	Ash %	Calories (kcal g ⁻¹)	Total %
Day 0	18	78.3	15.6	1.82	1.29	1.04	97.1
Day 14	19	75.7	17.8	2.70	1.47	1.23	97.6
Day 28	20	74.9	14.9	3.51	1.40	1.14	94.7
Day 56	21	75.5	17.0	3.27	1.53	1.24	97.3
Day 112	23	75.7	13.4	3.71	1.58	1.07	94.5
Day 263	31	78.5	15.2	2.29	1.21	1.05	97.1

6.8 Liver microsomal mixed function oxygenases

Liver microsomal mixed function oxygenases were measured in several sets of burbot (Table 16), and other species (Table 17). Comparison of the three northern sets of burbot enzyme activities showed a range of mean AHH activities from 0.008 to 0.046 nmol/mg prot./min, while the Lake Winnipeg group had a mean of 0.012. The 'control' samples fell within the range of the northern samples, and so it could be concluded that there was no AHH induction in the northern burbot. However, the interpretation of the enzyme assays may be complicated by differences in the organochlorine contamination between the Lake Winnipeg and northern samples. Burbot from Lake Winnipeg had higher levels of DDT and PCBs than the northern fish, but the northern burbot had as high or higher levels of toxaphene and chlordane (see sec. 5.11 below). PCBs contain congeners that induce the same pattern of microsomal oxidases as petroleum hydrocarbons, and so the Lake Winnipeg burbot may not have provided an adequate 'control' for comparison with the northern burbot in terms of microsomal enzyme activities. That is, it may be argued that the Lake Winnipeg burbot should have higher levels of MFO activities due to their higher levels of PCB contamination, hence elevated levels in the 'controls' would mean that comparable elevations in the northern burbot would not be recognizable. If there was any induction in the northern burbot, then it must have been smaller in amount than any hypothetical induction by PCBs in the fish from Lake Winnipeg.

The ages of the fish in the different groups also differed, and some of the variation may reflect biological variables like age or genetic strain. The relatively large variation found among strains of

Table 16: Liver microsomal oxidase enzyme activities (nanomoles product formed per mg microsomal protein per min) in burbot captured wild at several locations.

Lot 3, Fort Good Hope, April, 1985, N=13

	AHH	EROD	ODM	BZND	APND	P-450
Mean	.046	.052	.033	-	-	-
S.D.	.0193	.0326	.0230	-	-	-

Lot 10, Lake Winnipeg, February, 1986, N=25

	AHH	EROD	ODM	BZND	APND	P-450
Mean	.012	.042	.013	-	-	-
S.D.	.0079	.0436	.0112	-	-	-

Lot 15, Fort Good Hope, September, 1986, N=28

	AHH	EROD	ODM	BZND	APND	P-450
Mean	.008	.029	.030	1.65	1.58	.208
S.D.	.0044	.0152	.0151	.7172	.8329	.0573

Lot 16, Peel River, October, 1986, N=6

	AHH	EROD	ODM	BZND	APND	P-450
Mean	.0195	.0700	.0472	0.750	0.8333	.166
S.D.	.0101	.0514	.0162	0.5541	0.5989	.068

AHH — Aryl hydrocarbon hydroxylase
EROD — Ethoxyresorufin-O-deethylase
ODM — *p*-Nitroanisole-O-demethylase
BZND — Benzphetamine-N-demethylase
APND — Aminopyrine-N-demethylase
P-450 — Cytochrome P-450

Table 17: Liver microsomal oxidase enzyme activities (nanomoles product formed per mg microsomal protein per min) for catches (with $N \geq 6$) of species other than burbot captured at several northern locations.

Lake whitefish, Lot 16, Peel River, October, 1986, N=6						
	AHH	EROD	ODM	BZND	APND	P-450
Mean	.064	.074	.110	-	1.767	.197
S.D.	.215	.048	.012	-	.356	.045

Broad whitefish, Lot 15, Fort Good Hope, September, 1986, N=13						
	AHH	EROD	ODM	BZND	APND	P-450
Mean	.016	.005	.124	2.59	1.562	.141
S.D.	.012	.003	.077	1.85	.733	.066

Arctic cisco, Lot 15, Fort Good Hope, September, 1986, N=15						
	AHH	EROD	ODM	BZND	APND	P-450
Mean	.0176	.0127	-	-	1.720	.156
S.D.	.0131	.0115	-	-	.782	.071

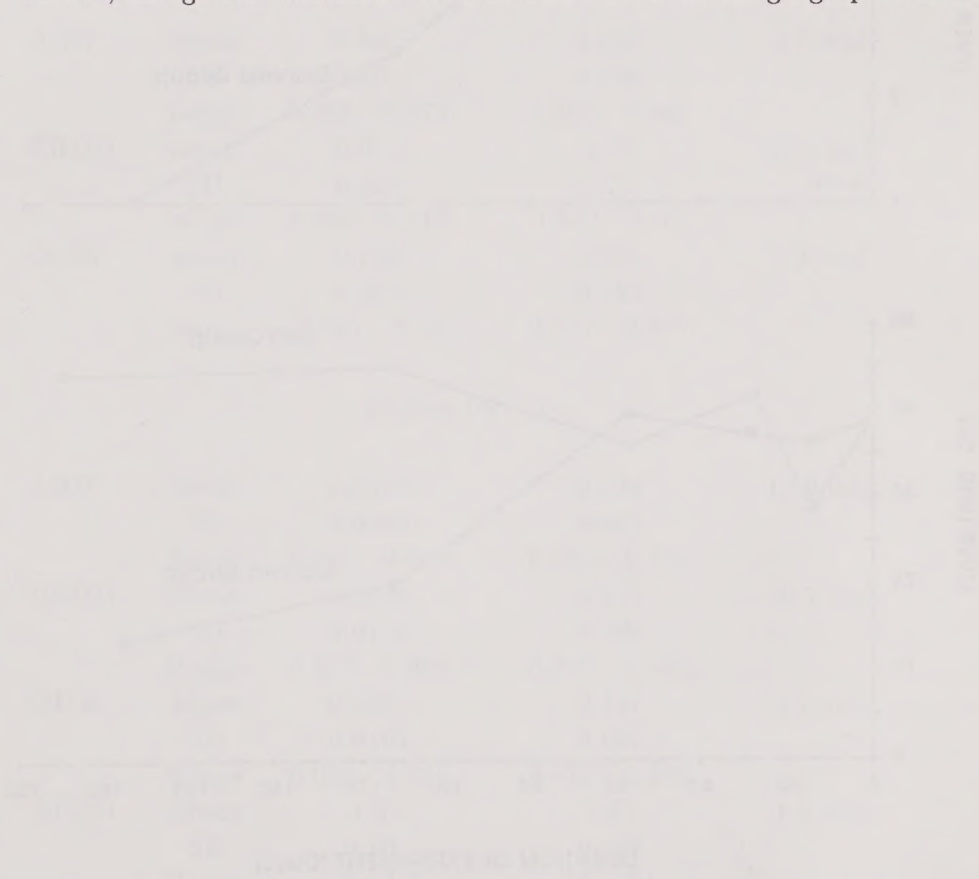
Inconnu, Lot 15, Captured at Fort Good Hope, September, 1986, N=6						
	AHH	EROD	ODM	BZND	APND	P-450
Mean	.008	.025	-	-	1.867	.247
S.D.	.003	.009	-	-	.314	.051

rainbow trout [124] was not apparent in the activities reported here. Seasonal variation in fish MFO enzyme levels has been reported [94], and that may have contributed to some of the differences among fish taken at different seasons, but it must have been relatively small, since the entire range was only about 5-fold.

Table 18 gives the results of experiments in which arctic charr and burbot were treated with Norman Wells crude oil, and MFO enzyme activities (EROD, AHH, ODM) were enhanced relative to controls. The most striking response was in EROD activity for which the difference between treated and untreated fish was about 40-fold, while AHH and ODM activities increased about 9- and 6-fold respectively. A similar experiment in which burbot were treated with oil or several other potential MFO inducing compounds is summarized in Table 19 (from Lockhart and Muir [86]). It is clear from these experiments that Norman Wells crude oil included compounds capable of potent induction of burbot MFO activities, however, toxaphene, chlordane and phenobarbital did not.

Previous studies with oil have established that petroleum exposure induces enhanced activities of AHH [166], and this activity has been used to monitor the response of fish to oil pollution

[121] and to detect subtle changes in fish around oil-production activity in the North Sea [26]. The authors are not aware of previous study on the induction of ODE or EROD by whole oil, although the polycyclic aromatic hydrocarbon 3-methylcholanthrene has been shown to induce these activities [41, 58]. All three activities in fish (charr and burbot, Tables 18 and 19) and marine mammals (seal pups) responded to oil treatments (Lockhart and Metner, unpublished). Based on the pattern of responses established in mammals and fish, [78] the pattern observed in Fort Good Hope burbot is not that expected from fish suffering exposure to oil. With regard to several other inducers, several species of fish have failed to demonstrate the mammalian type of induction by compounds like DDT and phenobarbital [1, 59, 164]. Toxaphene induced AHH weakly in channel catfish under some dietary regimens [98] but it did not do so in the experiments reported here. The mean EROD activities plotted in Figure 5 show the similarity between northern and southern populations of burbot, and give no indication of a difference between the geographic areas.



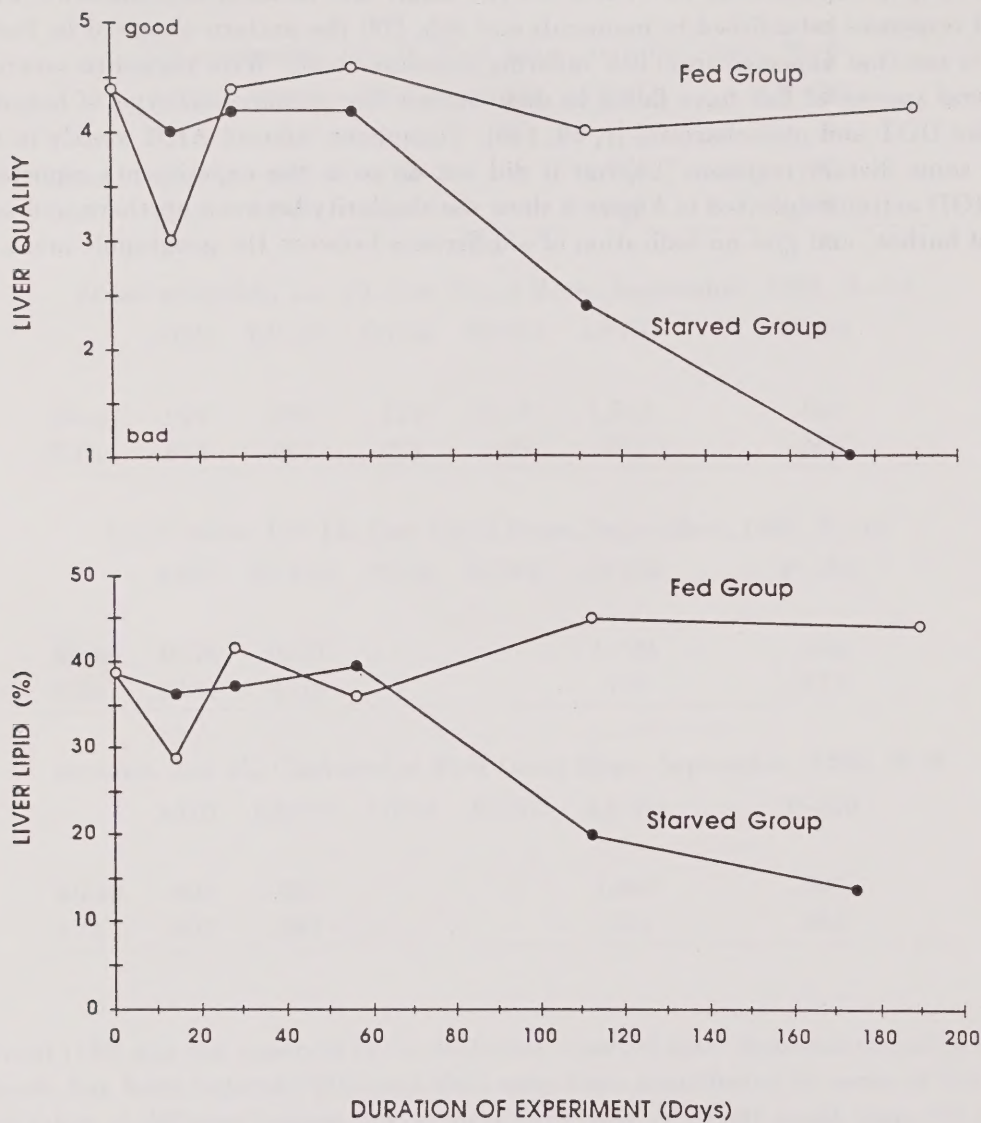


Figure 4: Liver quality ratings (arbitrary scale of 1 = bad to 5 = good) (top) and total liver lipid content (%) (bottom) in burbot subjected to starvation for 6 months in the laboratory.

Table 18: Liver microsomal enzyme induction in arctic charr and burbot treated with Norman Wells crude oil (nanomoles product formed per mg microsomal protein per min).

Enzyme Activity		Controls (corn oil only)	Oil-treated fish (petroleum oil and corn oil)	Induction
Arctic Charr (N=4)				
AHH	mean	0.062	0.550	8.9 fold
	SD	0.01	0.098	
	range	0.056 - 0.076	0.491 - 0.668	
EROD	mean	0.082	3.47	42.3 fold
	SD	0.024	1.71	
	range	0.060 - 0.113	1.921 - 5.91	
ODM	mean	0.056	0.322	5.8 fold
	SD	0.039	0.143	
	range	<0.001 - 0.088	0.167 - 0.497	
Burbot (N=3)				
AHH	Mean	0.0073	0.094	12.9 fold
	SD	0.0059	0.053	
	Range	0.003 - 0.014	0.035 - 0.138	
EROD	Mean	0.0300	0.621	20.7 fold
	SD	0.0114	0.306	
	Range	0.022 - 0.043	0.268 - 0.809	
ODM	Mean	0.0357	0.147	4.1 fold
	SD	0.0107	0.067	
	Range	0.024 - 0.045	0.073 - 0.202	
APND	Mean	1.3	1.67	1.3 fold
	SD	0.10	0.289	
	Range	1.2 - 1.4	1.5 - 2.0	
BZND	Mean	1.3	1.567	1.2 fold
	SD	0.436	0.513	
	Range	0.8 - 1.6	1.0 - 2.0	
P-450	Mean	0.278	0.295	1.1 fold
	SD	0.0125	0.0797	
	Range	0.268 - 0.292	0.247 - 0.387	

Activities defined as in Table 16

Table 19: Induction of burbot liver microsomal enzyme activities following intraperitoneal injections of corn oil or candidate inducing chemicals.

INDUCER	Mean Enzyme activity (nmol/mg protein/min at 25°C)					
	AHH			EROD		
	N	Mean	S.D.	N	Mean	S.D.
Corn oil 1	7	.009	.0037	7	.066	.031
Corn oil 2	6	.010	.0024	7	.071	.026
Corn oil 3	7	.0061	.0037	7	.062	.032
Benz(a)pyrene	6	.31	.088	6	2.72	.937
Crude oil	6	.206	.036	7	1.23	.204
Phenobarbital	7	.0054	.0035	7	.047	.026
Chlordane	7	.0063	.0028	7	.056	.028
Toxaphene	7	.0071	.0031	7	.048	.024

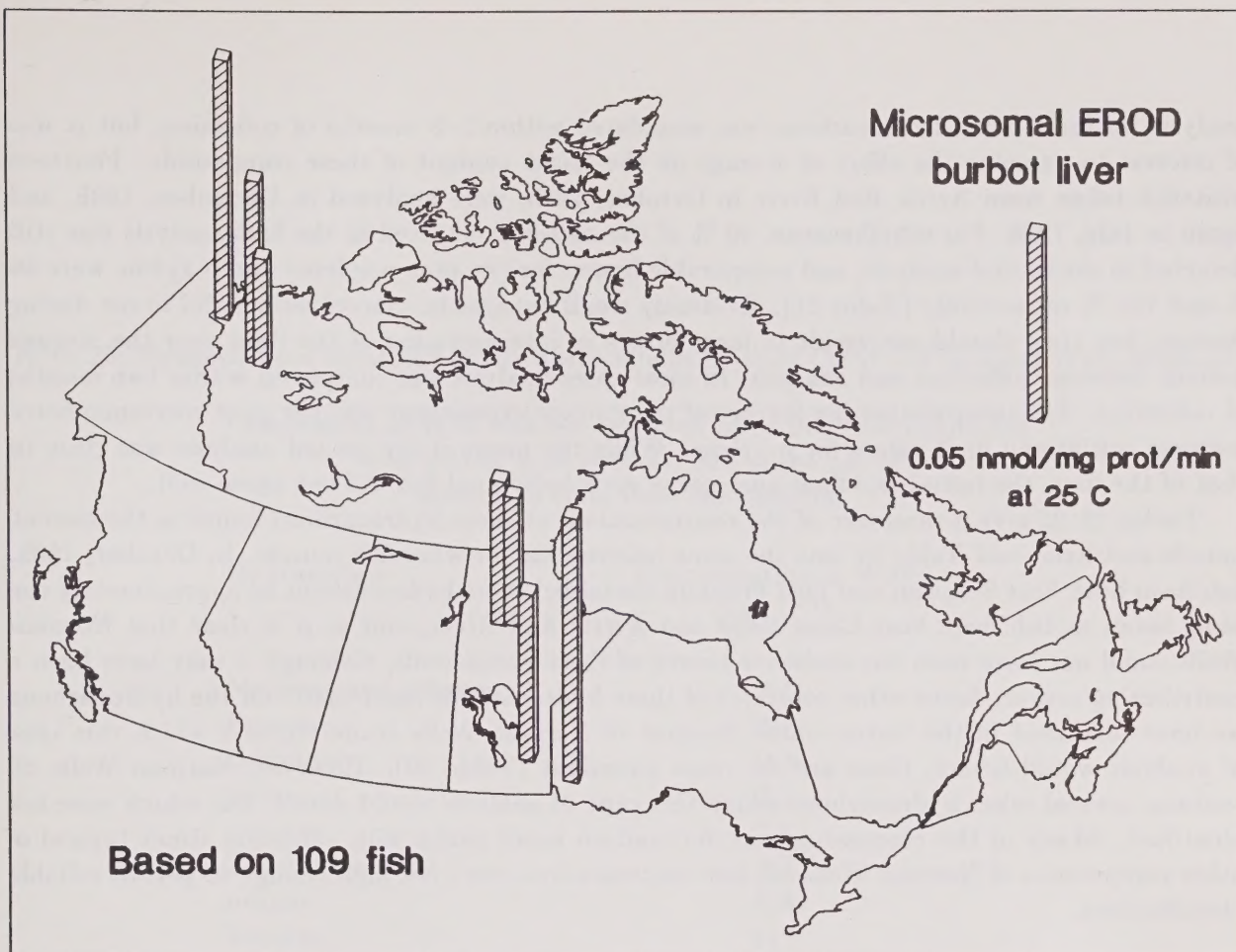


Figure 5: Liver microsomal EROD activities (nmoles product formed per mg microsomal protein per minute at 25°C) in burbot from several locations.

In view of the presence of organochlorines in all groups of burbot examined to date, probably the only ways to determine whether any given group had been induced is to compare its activity with and without an inhibitor specific to the induced form of the enzyme. However, even this might not establish whether the induction was caused by oil. Additional northern fish from drainages not likely to have experienced oil but with comparable residues of organochlorines will still be needed.

6.9 Low-boiling hydrocarbons in fish tissues

Murray *et al.* [109] identified twenty six hydrocarbons in the water-soluble fraction of Norman Wells crude oil (Table 20). The analysis used in this study would be expected to detect sixteen of these if they were present in fish tissue at concentrations in the low parts per billion (ng g^{-1}) range. Only four of these compounds were in fact identified, namely ethylbenzene, *m*- and *p*-xylene, and *o*-xylene. A representative chromatogram of a fish extract is shown in Figure 6. Peaks for *m*-xylene and *p*-xylene were not separated by the column, and so are both found as a single peak which might represent any mixture of the two isomers. The early samples from April, 1985, were analyzed using a more rapid temperature program than with subsequent samples, and resolution of *o*-xylene from another peak (heptanal) was not adequate for consistent quantitation of *o*-xylene in those early samples.

With relatively large numbers of samples arriving at the same time it was inevitable that some samples remained in the freezer for longer than others while awaiting analysis. Generally the

analysis for low-boiling hydrocarbons was completed within 2-3 months of collection, but it was of interest to examine the effect of storage on the tissue content of these compounds. Fourteen whitefish taken from Arctic Red River in October, 1985, were analyzed in December, 1985, and again in July, 1986. For ethylbenzene, 80 % of the material detected in the first analysis was still detected in the second analysis, and comparable figures for (*m*- plus *p*-xylene) and *o*-xylene were 66 % and 111 % respectively (Table 21). Evidently small changes in concentrations did occur during storage, but they should not result in large errors in interpretation of the data over the storage periods between collection and analysis. In most cases analysis was completed within two months of collection. The most distressing feature of the storage experiment was the poor correspondence between initial and final values for *o*-xylene. While the mean of the second analysis was close to that of the first, the initial and final analyses of each individual fish did not agree well.

Tables 22-23 give a summary of the concentrations of these hydrocarbons found in the burbot muscle and liver, and Table 24 lists the same information for whitefish muscle. In October, 1985, fish from both Fort Simpson and Fort Franklin contained these hydrocarbons at approximately the same levels as fish from Fort Good Hope and Arctic Red River, and so it is clear that Norman Wells could not have been the exclusive source of these compounds, although it may have been a contributing source. Some other source(s) of these hydrocarbons must exist. Of the hydrocarbons we have identified in the water-soluble fraction of Norman Wells crude oil, and which this type of analysis would detect, these are the most abundant (Table 20). However, Norman Wells oil contains several other hydrocarbons which this type of analysis would detect, but which were not identified. Many of the chromatograms did contain small peaks with retention times typical of other components of Norman Wells oil, but concentrations were not high enough to permit reliable identification.

Table 20: Hydrocarbons identified in the water-soluble fraction (WSF) of Norman Wells crude oil¹

Compounds in WSF but not expected to be detected in the fish even if they were present, due to the use of solvent to elute from the charcoal	
Hydrocarbon	Concentration in WSF (mg L ⁻¹)
Methane and ethane	9.1
Propane	2.7
Isobutane	11.5
butane	2.1
pentane	3.4
2-methylpentane	0.5
hexane	0.8
benzene	14.1
toluene	7.5
Compounds in WSF and expected to be detected if present in the fish above detection limits of about 1 ng g ⁻¹	
Hydrocarbon	Concentration in WSF (mg L ⁻¹)
ethylbenzene	0.7
<i>m</i> - and <i>p</i> -xylene	2.4
<i>o</i> -xylene	1.0
isopropylbenzene	0.1
<i>n</i> -propylbenzene	0.1
six C3 benzenes	(sum of 6) 1.7
two C4 benzenes	(sum of 2) 0.3
naphthalene	0.1
2-methylnaphthalene	0.1
1-methylnaphthalene	0.1

¹From Murray, Lockhart and Webster [109].

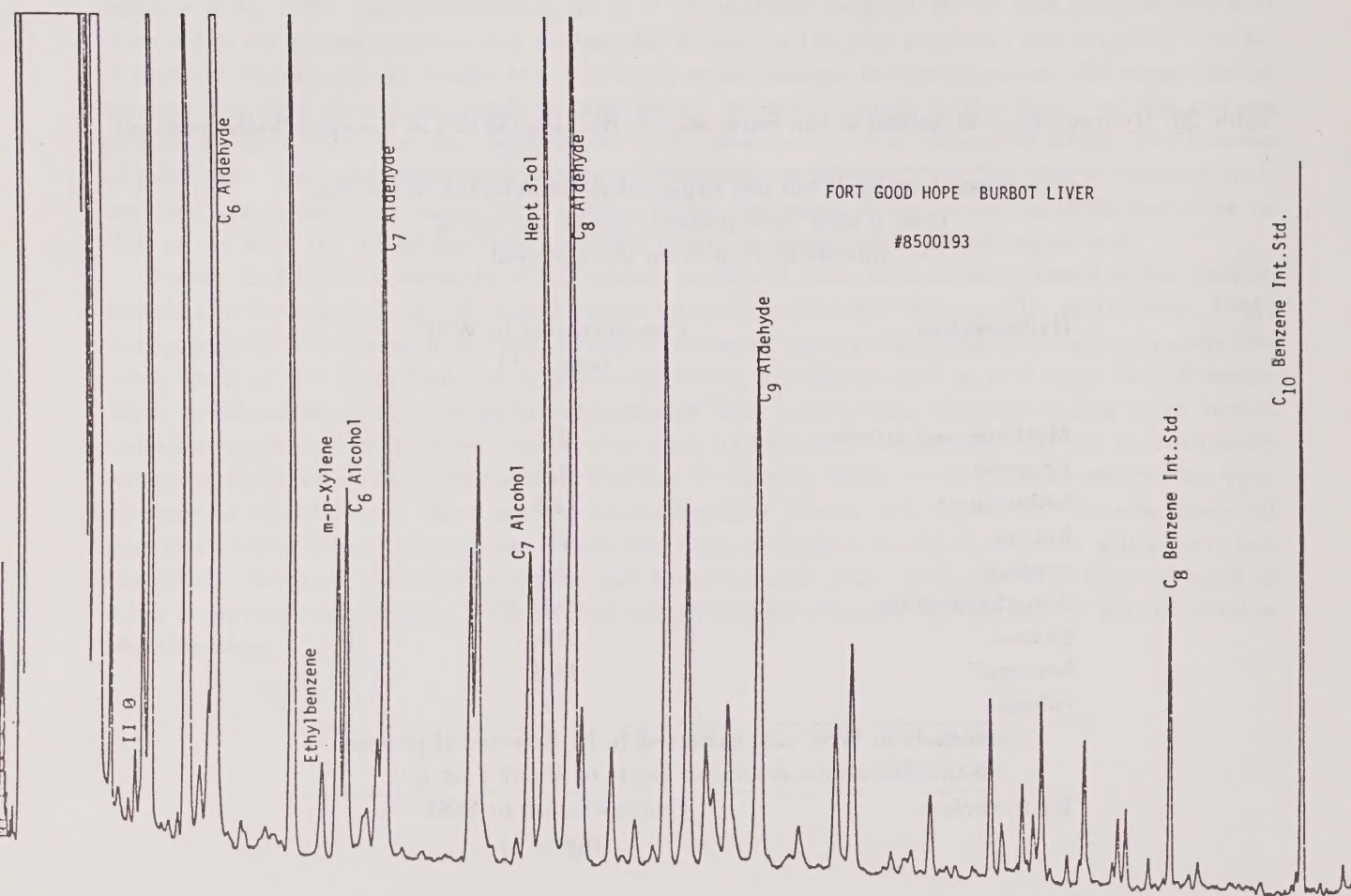


Figure 6: Representative gas chromatogram of volatile organic compounds (flame ionization detector) extracted from fish muscle by purging for 3 hours with air at 60°C, with identifications of peaks as indicated.

Table 21: Stability of ethylbenzene and xylenes (ng g^{-1}) in fourteen samples of muscle of whitefish during storage in a freezer. Samples were analyzed originally during December, 1985, and reanalyzed during July, 1986.

	Analyzed Dec., 1985			Analyzed July, 1986			Percent of original		
Number	ebenz	<i>m,p</i> -xy	<i>o</i> -xy	ebenz	<i>m,p</i> -xy	<i>o</i> -xy	ebenz	<i>m,p</i> -xy	<i>o</i> -xy
8500100	9.5	27.5	8.5	13.0	32.0	22.0	137	116	259
8500101	8.0	29.5	9.0	9.5	27.5	21.0	119	93	233
8500102	10.0	27.0	5.5	14.0	36.5	11.0	140	135	200
8500103	13.0	41.5	11.5	10.0	23.0	4.0	77	55	35
8500104	5.5	15.0	5.0	9.0	21.0	3.5	164	140	70
8500105	31.5	106.	17.0	11.0	23.5	3.0	35	22	18
8500106	nd	10.5	2.0	6.0	19.0	10.0	—	181	500
8500107	8.5	28.5	2.0	4.5	13.0	3.5	53	46	175
8500108	5.5	57.5	7.5	7.5	26.0	7.5	136	45	100
8500109	12.5	32.0	3.0	6.5	26.5	8.0	52	83	267
8500111	14.0	47.5	5.5	6.5	21.0	3.5	46	44	64
8500112	8.0	27.0	9.0	7.0	28.0	17.0	88	104	189
8500115	12.0	45.5	12.5	6.5	27.0	nd	54	59	0
8500116	32.0	125.	15.5	24.5	88.0	12.5	77	70	81
Mean	12.1	44.3	8.1	9.68	29.4	9.03	80.0	66.4	111
SD	9.06	32.8	4.7	5.07	17.8	6.96			

ebenz = ethylbenzene

m,p-xy = *m*- and *p*-xylenes

o-xy = *o*-xylene

nd = not detected

Table 22: Ethylbenzene and xylenes (ng g⁻¹) detected in burbot muscle tissue.

Source of fish	Compound	mean	SD	minimum	maximum	N
Fort Good Hope	Ethylbenzene	49.6	26.4	20.0	115.	16
April, 1985, Lot 3	<i>m</i> -, <i>p</i> -xylene	185.	110.	72.0	499.	16
Fort Good Hope	Ethylbenzene	7.34	6.07	1.5	25.0	16
October, 1985,	<i>m</i> -, <i>p</i> -xylene	15.9	10.2	4.0	46	
Lot 7	<i>o</i> -xylene	0.47	1.15	nd	4.0	16
Fort Good Hope	Toluene	0.67	1.04	nd	5.0	45
September, 1986	Ethylbenzene	2.45	2.02	nd	7.5	45
Lot 15	<i>m</i> -, <i>p</i> -xylene	11.1	20.8	1.0	146	45
	<i>o</i> -xylene	0.2	0.5	nd	1.5	45
Fort Franklin	Ethylbenzene	12.2	5.39	4.0	29.0	21
October, 1985	<i>m</i> -, <i>p</i> -xylene	35.7	17.8	12.5	75.0	21
Lot 6	<i>o</i> -xylene	0.55	1.05	nd	3.5	21
Arctic Red River	Ethylbenzene	7.0	5.59	nd	20.0	22
October, 1985	<i>m</i> -, <i>p</i> -xylene	16.7	13.9	nd	57.5	22
Lot 5	<i>o</i> -xylene	0.21	0.61	nd	2.5	22
Fort Simpson	Ethylbenzene	7.78	4.02	2.0	19.0	20
October, 1985	<i>m</i> -, <i>p</i> -xylene	21.2	12.5	7.0	57.0	20
Lot 8	<i>o</i> -xylene	0.20	0.50	nd	1.50	20
Fort Simpson	Ethylbenzene	4.75	4.82	nd	12.5	8
April, 1986	<i>m</i> -, <i>p</i> -xylene	14.9	13.3	nd	34.0	8
Lot 12	<i>o</i> -xylene	0.13	0.35	nd	1.0	8
Great Slave Lake	Ethylbenzene	7.77	6.61	1.0	26.0	13
April, 1985, Lot 4	<i>m</i> -, <i>p</i> -xylene	13.7	7.08	5.0	33.5	13
Lake Winnipeg	Ethylbenzene	5.70	5.14	nd	12.0	5
February, 1986	<i>m</i> -, <i>p</i> -xylene	20.6	13.0	6.0	34.5	5
Lot 10	<i>o</i> -xylene	nd	—	nd	nd	5

Table 23: Ethylbenzene and xylenes (ng g⁻¹) detected in burbot liver tissue.

Source of fish	Compound	mean	SD	minimum	maximum	N
Fort Good Hope	Ethylbenzene	33.4	12.8	19.5	51.0	7
October, 1985	<i>m</i> -, <i>p</i> -xylene	124.	55.7	58.0	218.	7
Lot 7	<i>o</i> -xylene	18.2	17.4	1.5	54.0	7
Fort Franklin	Ethylbenzene	39.6	23.2	11.0	84.0	8
October, 1985	<i>m</i> -, <i>p</i> -xylene	133.	90.3	17.0	321.	8
Lot 6	<i>o</i> -xylene	76.8	82.8	11.0	256.	8
Arctic Red River	Ethylbenzene	46.3	—	25.5	67.0	2
October, 1985	<i>m</i> -, <i>p</i> -xylene	144.	—	97.5	190.	2
Lot 5	<i>o</i> -xylene	21.3	—	15.5	27.0	2
Fort Simpson	Ethylbenzene	22.9	18.6	nd	54.5	17
October, 1985	<i>m</i> -, <i>p</i> -xylene	81.9	65.3	4.0	188.	17
Lot 8	<i>o</i> -xylene	9.71	11.4	nd	42.0	17
Fort Simpson	Ethylbenzene	1.81	2.17	nd	5.5	8
April, 1986	<i>m</i> -, <i>p</i> -xylene	4.44	5.53	nd	16.0	8
Lot 12	<i>o</i> -xylene	0.63	1.19	nd	3.0	8
Fort Good Hope	Toluene	9.9	5.56	4.0	26.0	15
September, 1986	Ethylbenzene	31.8	13.6	14.5	66.5	15
Lot 15	<i>m</i> -, <i>p</i> -xylene	113.6	52.2	54.5	238.5	15
	<i>o</i> -xylene	24.3	14.5	5.5	49.0	15

A map showing the levels of *m*- and *p*-xylenes in burbot muscle is shown in Figure 7. Only one northern sample was elevated relative to the remaining northern and southern samples, and it was the one taken under the ice in April, 1985.

Preliminary experimental exposure of fish (arctic charr) to radiolabelled *o*-xylene showed that while this hydrocarbon was readily taken up into body tissues during six-hr exposures, it fell rapidly after the fish were transferred to clean water. With these young charr a half-life of 6-7 hr was indicated for total radioactivity in the fish at temperatures of both 5°C and 15°C (Table 25). Presumably this reflects rapid metabolism of these hydrocarbons with excretion of metabolites, possibly via the bile as described by Krahn *et al.* [75] for fish from Puget Sound. Niimi and Palazzo [114] have recently determined the persistence of several PAH compounds in rainbow trout, and found that all were cleared readily from the fish with half-lives of nine days or less. The implication of rapid metabolism and clearance is two-fold, namely that exposure of the fish in the Dene communities was recent, and also that measurement of residues of parent unmetabolized hydrocarbons is an underestimate of hydrocarbon intake.

Table 24: Ethylbenzene and xylenes (ng g⁻¹) detected in whitefish muscle tissue.

Source of fish	Compound	mean	SD	minimum	maximum	N
Broad whitefish	Ethylbenzene	16.1	11.1	nd	39.0	25
Fort Good Hope	<i>m</i> -, <i>p</i> -xylene	70.8	46.6	3.0	173.	25
October, 1985, Lot 7	<i>o</i> -xylene	9.26	8.08	nd	26.0	25
Broad whitefish	Toluene	3.96	1.61	2.0	7.0	13
Fort Good Hope	Ethylbenzene	7.46	4.76	1.5	20.5	13
September, 1986	<i>m</i> -, <i>p</i> -xylene	29.2	18.0	10.5	76.5	13
Lot 15	<i>o</i> -xylene	10.8	10.7	1.0	38.0	13
Lake whitefish	Ethylbenzene	104.	79.5	34.0	273.	7
Fort Good Hope	<i>m</i> -, <i>p</i> -xylene	406.	339.	114.	1122.	7
April, 1985, Lot 3						
Lake whitefish	Ethylbenzene	14.7	6.15	6.0	29.0	16
Fort Good Hope	<i>m</i> -, <i>p</i> -xylene	59.0	25.3	26.0	118.	16
October, 1985, Lot 7	<i>o</i> -xylene	3.0	1.82	nd	6.0	16
Lake whitefish	Ethylbenzene	13.7	9.82	4.0	43.5	19
Fort Franklin	<i>m</i> -, <i>p</i> -xylene	48.2	36.5	6.0	157.	19
October, 1985, Lot 6	<i>o</i> -xylene	12.6	11.0	2.0	47.5	19
Lake whitefish	Ethylbenzene	12.9	7.80	5.5	32.0	16
Arctic Red River	<i>m</i> -, <i>p</i> -xylene	44.7	29.5	15.0	125.	16
October, 1985, Lot 5	<i>o</i> -xylene	9.53	5.97	2.0	24.5	16
Lake whitefish	Ethylbenzene	24.3	17.4	nd	74.0	19
Fort Simpson	<i>m</i> -, <i>p</i> -xylene	83.0	63.4	9.5	266.	19
October, 1985, Lot 8	<i>o</i> -xylene	2.5	4.06	nd	12.5	19
Lake whitefish	Ethylbenzene	10.6	6.93	1.0	23.	12
Northern Manitoba	<i>m</i> -, <i>p</i> -xylene	47.2	35.0	6.0	110.	12
1986, Lot 13	<i>o</i> -xylene	21.7	16.2	3.0	46.	12

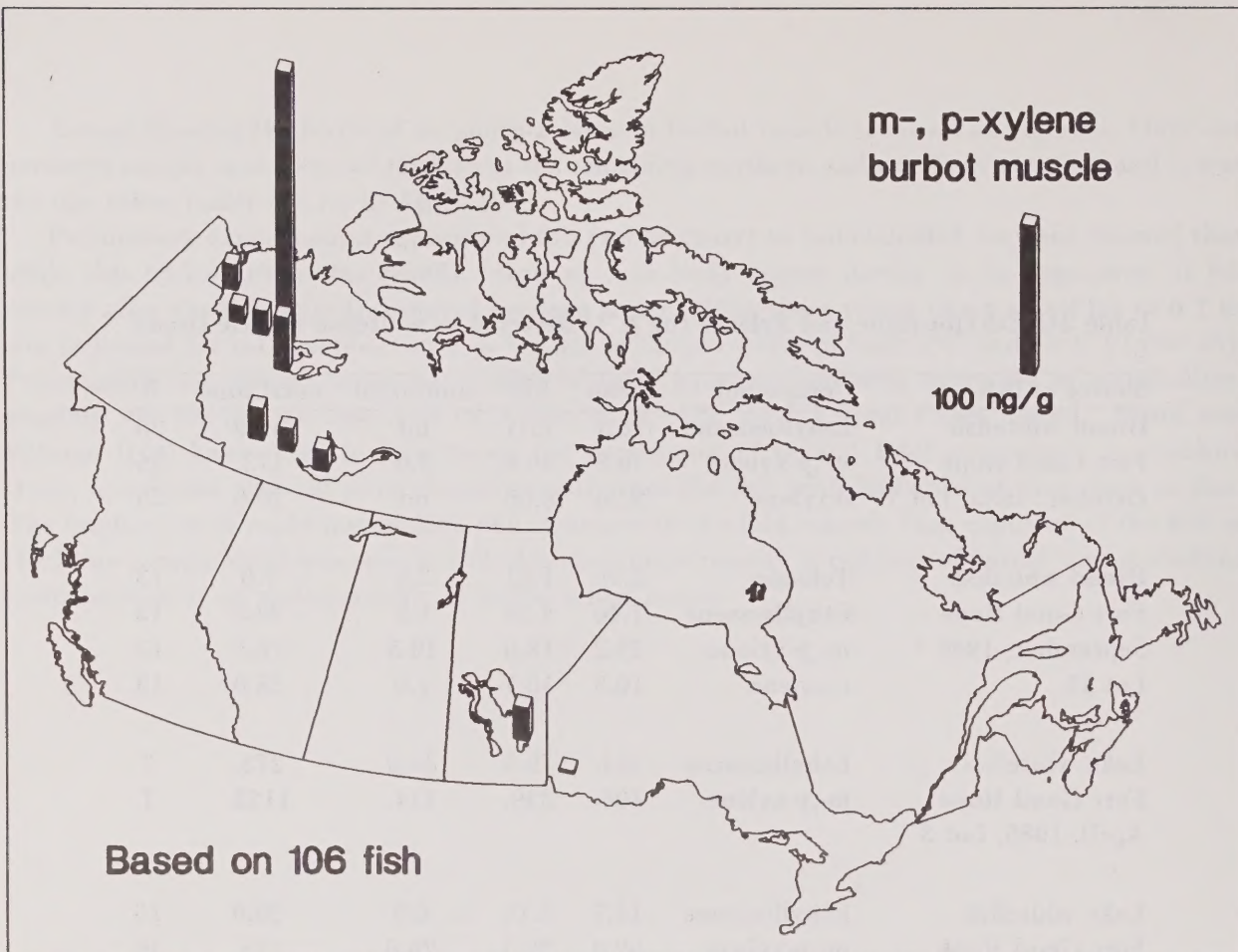


Figure 7: Map showing xylenes in muscle of burbot from several locations

One probable explanation for widespread recent exposure of fish to hydrocarbons would be a general dispersal of hydrocarbons through the air. Aerial dispersal of low-boiling petroleum hydrocarbons has been implicated previously [46, 67, 68], and it has been shown also for higher-boiling polyaromatic hydrocarbons which are transported adsorbed to fine particles in air [25].

Some of the results may bear on the question of sources. If the source were strictly aerial, then one might expect low levels in the fish in late winter because inputs should be restricted by cover of ice and snow. On the other hand, if the source were within the river, then one might expect high levels in fish in late winter because cover by ice and snow would restrict evaporative loss of low-boiling materials. An example of this limitation in evaporative loss of a light oil occurred following the Cameron River spill of diesel fuel [83]. Three sets of analyses bear on these arguments. Comparing the hydrocarbon content of fish from Fort Good Hope in April, 1985, with that in October, 1985, (Tables 22 and 23, burbot; Table 24, whitefish) there was approximately a six-fold difference with the late winter (ice cover) sample higher than the late summer (open water) sample. This suggests a source beneath the ice at some location from which hydrocarbons could become available to fish at Fort Good Hope. However, comparing burbot from Fort Simpson in October, 1985, with those from April, 1986, the corresponding increase in late winter was not detected, which suggests that the river source indicated at Fort Good Hope was not operating at Fort Simpson. Taken together these results suggest a source of hydrocarbons somewhere within the river between Fort Simpson and Fort Good Hope (presumably from seepage and/or effluents at Norman Wells) superimposed on top a more general background, possibly contributed by dispersal of hydrocarbons from distant sources by air movements. The mean levels of ethylbenzene and *m*-, *p*-xylene in September, 1986, were much lower than they were in both burbot and broad whitefish during the previous fall.

Low-boiling hydrocarbons have been found in earlier analyses of water and fish. Strachan and Edwards [151] cited an Ontario Ministry of Environment report on pollutants in water at Niagara-on-the-Lake, in which xylenes were reported at $0.16 \mu\text{g L}^{-1}$. Slater and Blok [146] identified a number of hydrocarbons (benzene, alkylated benzenes, alkanes, naphthalene, and others) in the water of Buffalo Pound Lake (Sask.) during their investigations of algal blooms, but the source(s) of the compounds was not identified unequivocally. Reinert, Hunter and Sabatino [135] examined a small number of estuarine fish and shellfish from New Jersey and found up to 0.18 ppm (180 ng g^{-1}) of *m*- and *p*-xylene in them. Berg [8] identified '*p*-cymene and other alkyl- and alkenyl benzenes' in Norwegian salmon from rivers polluted with paper mill effluents, but concentrations were not given. Ogata and Miyake [117, 118] associated an objectionable odour in fish from the coast of Japan with contamination by toluene. Josephson and co-workers [63, 64, 65, 66] have identified alkylated benzenes in several species of fish, mostly from Wisconsin waters, with no known history of contamination. For example, the concentration of 1,3-dimethylbenzene (*m*-xylene) in emerald shiners ranged from about 5 to 8 ng g^{-1} , depending on the method of extraction, but this compound was not reported in 'slime' from fresh lake whitefish from Lake Michigan, although ethenylbenzene and trimethylbenzene were found at about 1 and 3 ng g^{-1} respectively. Interestingly, 1,2-dimethylbenzene (*o*-xylene) was not reported in slime from fresh whitefish [63], but it was found in slime from whitefish held frozen in storage for over a year [66]. From the reports cited above it seems that the compounds being identified in northern fish are unusual neither in kind nor in quantity. These hydrocarbons have been found in fish before and the concentrations found in this study are within ranges defined by previous studies. However, the question of the sources of the low boiling hydrocarbons in northern fish is still open, particularly for fish taken late in the winter season.

In addition to the substituted benzenes mentioned above, a number of other materials were present in the muscle extracts, and these have been identified tentatively from mass spectra mostly

Table 25: Uptake and clearance of radiolabelled *o*-xylene by young arctic charr at temperatures of 5°C and 15°C.

Temperature		5°C	15°C
Mean fish weight (g)		1.365	1.561
Exposure concentration ($\mu\text{g mL}^{-1}$)	start (0 hr)	6.52	4.06
	end (6 hr)	5.10	2.22
Time in clean water		Concentration in fish ($\mu\text{g g}^{-1}$)	
0 hr		94.2	23.2
		187.7	58.6
		170.8	33.0
		142.2	42.4
23 hr		4.5	0.44
		6.7	0.84
		1.8	0.53
		2.0	0.68
45 hr		0.84	0.16
		1.17	0.35
		0.45	0.13
		2.12	0.41
Calculated half-life in fish		6.2 hr	6.1 hr

Table 26: Cancer related biological activities associated with individual polycyclic aromatic hydrocarbons¹

Compound	Biological Activity
benz(<i>a</i>)anthracene	tumor initiator
chrysene	tumor initiator
Indeno(1,2,3- <i>c,d</i>)pyrene	tumor initiator
Benzo(<i>a</i>)pyrene	Complete carcinogen, tumor initiator
Benzo(<i>b</i>)fluoranthene	Complete carcinogen, tumor initiator
Dibenz(<i>a,h</i>)anthracene	Complete carcinogen, tumor initiator
Fluoranthene	Co-carcinogen with B(<i>a</i>)P
Pyrene	Co-carcinogen with B(<i>a</i>)P
Benzo(<i>g,h,i</i>)perylene	Co-carcinogen with B(<i>a</i>)P

¹From Gammage [44]

as aliphatic and olefinic aldehydes and alcohols containing from six to nine carbon atoms. These and other volatile compounds have been reported in freshwater fish in the series of papers by Josephson *et al.* [63, 64, 65, 66], and they are excluded from the tables here because they are believed to be derived from natural decomposition of lipids rather than from contamination.

6.10 Polycyclic aromatic hydrocarbons

Polyaromatic hydrocarbons were found in many of the fish samples analyzed; results for burbot are summarized in Table 27. The compounds most frequently identified were naphthalene (over 35 % of samples), phenanthrene and chrysene/benz(*a*) anthracene (over 25 % of samples), and fluorene and fluoranthene (over 20 % of samples), followed by benz(*a*)pyrene (about 5 % of samples). Naphthalene was found at concentrations over 100 ng g⁻¹ in some samples of burbot liver, similar to 72 ng g⁻¹ reported in burbot liver from Finland [133] (Table 28). Fluorene levels were also over 100 ng g⁻¹ in some livers. The remaining PAHs were well below those reported for the Finnish burbot. Phenanthrene ranged up to about 6 ng g⁻¹, consistent with results reported for cisco in the southern Beaufort Sea (4-15 ng g⁻¹) [171]. Fluoranthene was detected frequently with most samples under 1 ng g⁻¹, but one sample reached almost 5 ng g⁻¹. Lawrence and Weber [77] (Table 29) found fluoranthene at up to 1.4 ng g⁻¹ in fish from Lake Ontario. Benz(*a*)pyrene was found in only a few samples, at concentrations up to 1.25 ng g⁻¹, somewhat higher than the Lake Ontario results. Both burbot liver and muscle contained PAH with liver having generally having both a greater frequency of positive results, and also higher concentrations than muscle. Rainio *et al.* [133] also found higher concentrations of PAH in liver than in muscle of Finnish burbot.

The reason for interest in PAH compounds is their association with cancer, and much research has been done on the levels of PAH in air, water, foods, and cigarette smoke. The biological activities associated with several of the PAH detected in burbot and whitefish are listed in Table 26 (from Gammage [44]).

PAH compounds occur in many common foods [40] and concentrations of most unaltered PAH

reported here for burbot and whitefish do not appear to represent an unusual hazard for consumption in relative to concentrations in some other common foods. For example, Lawrence and Weber [77] found benz(a)pyrene in char-broiled hamburgers at 1.5 to 4.0 ppb, apparently as a result of char-broiling since B(a)P was not detected in fried hamburgers. A report on PAH released by the Ontario Ministry of the Environment [2][page 151] listed B(a)P concentration ranges in several foods from 0 to 48.1 ng g⁻¹. For example, vegetable oils ranged from 0.9 to 15 ng g⁻¹, and cereals were from 0.2 to 4.1 ng g⁻¹. Smoking a package of cigarettes results in inhalation of about 1 µg of B(a)P [2][page 53]. If any difference between dietary and inhalation exposure is ignored, then taking the levels of B(a)P in liver (Table 27) as approximately 1 ng g⁻¹, consumption of about 1000 g of fish would furnish the same intake as smoking a package of cigarettes (1 µg of B(a)P).

Concern about cancer resulting from the exposure to PAH compounds extends to the fish themselves. In some cases fish populations have developed unusually high incidences of neoplasms [107, 13] and some of these cases these have been convincingly associated with exposure to PAHs [13, 75], apparently by accumulation of PAH from sediments through food [95]. The levels of PAH in bullheads with high incidences of tumors have been measured [7], and they are well above those found in northern burbot and whitefish. It is of interest that the control area used by Baumann, Smith and Ribick [7] was Buckeye Lake in Ohio, and the levels there were generally lower than those in northern fish. There are no reports of unusual incidences of tumors in Mackenzie River fish, and the one unusual structure found in a burbot liver from the April (1985) sample was submitted to the Smithsonian Institution and found to be the result of a mechanical injury. As in the case of low-boiling hydrocarbons, fish can metabolize PAH compounds and excrete them, generally as conjugates with sugar or other derivatives through the bile. This again implies that measurement of the parent unmetabolized PAH compounds gives a systematic underestimate of the intake and exposure of the fish to PAH. Metabolism of PAH by fish has been reviewed recently by Tan and Melius [153].

Table 27: Polyaromatic hydrocarbons in burbot liver and muscle from Arctic communities. (No. anal. = number analyzed. No. pos. = number above detection limits. Max. = maximum concentration found, in ng g⁻¹ wet weight.)

Tissue	Source	Lot		Nap	Acy	Bip	Ace	Flu	Phe	Ant	Fln	Pyr
Muscle	Ft G H	3	No. anal.	11	11	11	11	11	11	11	11	11
			No. pos.	4	0	0	0	0	0	0	1	0
			Max.	31.9	nd	nd	nd	nd	nd	nd	0.07	nd
Muscle	Grt Sl	4	No. anal.	13	13	13	13	13	13	13	13	13
			No. pos.	1	0	0	0	0	0	0	5	0
			Max.	5.38	nd	nd	nd	nd	nd	nd	0.08	nd
Liver	A R R	5	No. anal.	5	5	5	5	5	5	5	5	5
			No. pos.	2	0	0	0	3	1	1	0	1
			Max.	28.8	nd	nd	nd	40.8	1.58	2.10	nd	22.7
Liver	Ft Fr	6	No. anal.	3	3	3	3	3	3	3	3	3
			No. pos.	3	0	0	0	1	3	0	1	0
			Max.	108.	nd	nd	nd	18.4	3.15	nd	0.57	nd
Liver	Ft G H	7	No. anal.	4	4	4	4	4	4	4	4	4
			No. pos.	0	0	0	0	3	1	0	2	0
			Max.	nd	nd	nd	nd	32.6	0.96	nd	1.12	nd
Liver	Ft Sim	8	No. anal.	5	5	5	5	5	5	5	5	5
			No. pos.	4	0	0	0	3	4	0	2	0
			Max.	113.	nd	nd	nd	32.6	6.26	nd	2.18	nd
Liver	Ft G H	15	No. anal.	15	15	15	15	15	15	15	15	15
			No. pos.	13	0	0	0	11	10	0	2	0
			Max.	137.	nd	nd	nd	180.	4.47	nd	4.67	nd
All			No. anal.	97	97	97	97	97	97	97	97	97
			No. pos.	38	1	0	0	22	26	1	22	1
			Max.	137	13.7	nd	nd	180	6.26	2.1	4.67	22.7

Ft G H Fort Good Hope
A R R Arctic Red River
Ft Sim Fort Simpson

Grt Sl Great Slave Lake
Ft Fr Fort Franklin

Table 27: (Continued) Polyaromatic hydrocarbons in burbot liver and muscle from Arctic communities. (No. anal. = number analyzed. No. pos. = number above detection limits. Max. = maximum concentration found, in ng g⁻¹ wet weight.)

Tissue	Source	Lot		B/Chr	BbF	BkF	BaP	IdP	DbA	BPe
Muscle	Ft G H	3	No. anal.	11	11	11	11	11	11	11
			No. pos.	5	0	1	0	0	0	0
			Max.	2.59	nd	1.62	nd	nd	nd	nd
Muscle	Grt Sl	4	No. anal.	13	13	13	13	13	13	13
			No. pos.	0	0	0	0	0	1	0
			Max.	nd	nd	nd	nd	nd	0.10	nd
Liver	A R R		No. anal.	5	5	5	5	5	5	5
			No. pos.	1	0	0	0	0	0	0
			Max.	2.74	nd	nd	nd	nd	nd	nd
Liver	Ft Fr	6	No. anal.	3	3	3	3	3	3	3
			No. pos.	2	0	0	0	0	0	0
			Max.	1.34	nd	nd	nd	nd	nd	nd
Liver	Ft G H	7	No. anal.	4	4	4	4	4	4	4
			No. pos.	3	0	0	2	0	0	1
			Max.	2.68	nd	nd	1.25	nd	nd	0.14
Liver	Ft Sim	8	No. anal.	5	5	5	5	5	5	5
			No. pos.	3	0	0	2	0	1	0
			Max.	2.48	nd	nd	1.02	nd	0.48	nd
Liver	Ft G H	15	No. anal.	15	15	15	15	15	15	15
			No. pos.	8	0	0	1	0	0	0
			Max.	5.74	nd	nd	1.14	nd	nd	nd
All			No. anal.	97	97	97	97	97	97	97
			No. pos.	25	0	1	5	0	2	0
			Max.	5.74	nd	1.62	1.25	nd	0.48	nd

Nap - Naphthalene	Acy - Acenaphthylene	Bip - Biphenyl
Ace - Acenaphthene	Flu - Fluorene	Phe - Phenanthrene
Ant - Anthracene	Fln - Fluoranthene	Pyr - Pyrene
B/Chr - Benz(<i>a</i>)anthracene/Chrysene	BbF - Benz(<i>b</i>)fluoranthene	
BkF - Benz(<i>k</i>)fluoranthene	BaP - Benz(<i>a</i>)pyrene	
IdP - Indeno(1,2,3- <i>c,d</i>)pyrene	DbA - Dibenzo(<i>a,h</i>)anthracene	
Bpe - Benz(<i>g,h,i</i>)perylene		

Table 28: Published concentrations (ng g^{-1}) of some polycyclic aromatic hydrocarbons in burbot from the Finnish archipelago¹.

Tissue	Nap	Phe	Fln	Pyr	BaA/Chr Tri	Total PAH
Muscle	<0.5	14	5	4	3	26
Liver	72	86	45	62	180	445
Gall (bile)	215	44	29	25	—	313
Nap – naphthalene Phe – phenanthrene Fln – fluoranthene Pyr – pyrene BaA – benz(a)anthracene Chr – chrysene Tri – triphenylene						

¹From Rainio *et al.* [133]

Table 29: Published concentrations (ng g^{-1}) of some polycyclic aromatic hydrocarbons in fish from Lake Ontario¹.

PAH	white perch	crappies	smelt	smelt	eel	catfish
Fluoranthene	1.1	0.6	1.0	0.6	1.4	0.8
Pyrene	2.3	1.1	1.0	1.4	1.9	1.8
Benz(b)fluoranthene	0.1	—	—	—	0.2	0.2
Dimethylphenanthrene	0.5	0.1	0.1	0.2	0.3	0.3
Benz(a)anthracene	0.4	0.2	0.2	0.1	0.2	0.3
Perylene	—	—	—	—	—	0.1
Benz(a)pyrene	—	0.1	0.1	—	—	—
Dibenz(ac)anthracene	—	—	0.1	0.2	0.8	—
Dibenz(ah)anthracene	—	—	1.6	2.2	2.9	—
Picene	—	—	—	—	0.1	—
Indeno(1,2,3-c,d)pyrene	0.1	—	—	—	0.1	—

Dashes (—) indicate $<0.1 \text{ ng g}^{-1}$.

¹From Lawrence and Weber [77]

6.11 Organochlorines

Organochlorine compounds identified in the fish are listed in Table 31. A representative chromatogram is shown in Figure 8. All the samples contained organochlorines with toxaphene, PCBs, and chlordane at highest concentrations. There was a striking increase in the organochlorines present in the burbot liver as compared with whitefish muscle, presumably reflecting the higher fat content in the burbot liver. Burbot from Lake Winnipeg generally exceeded Arctic burbot by several fold for PCBs and DDT, but were comparable for the other organochlorines. Mean levels of chlordane and toxaphene were highly variable but were somewhat higher in livers of the Arctic burbot, although the difference was not statistically significant. It is difficult to compare organochlorines in burbot liver in this report with earlier publications on burbot (Table 32) because the previous data are so few and represent either whole fish or muscle. Burbot collected in the fall of 1974 from Lake Superior contained much more DDT, dieldrin and PCB (whole fish basis) [4] than the northern burbot. PCB levels in burbot liver from Churchill Falls, Labrador, were generally higher than the arctic burbot, but comparable with those from Lake Winnipeg [111]. Total DDT levels were up to 1.81 ppm in a group of Lake Superior fish with a fat content (hexane extractable material only) of 5 percent. Wong [172] summarized the limited organochlorine residue data available for commercial fish from the Canadian Arctic as determined by the Inspection Service of the Department of Fisheries and Oceans, and the maximum DDT concentration recorded was 30 ppb in whitefish from Hay River in 1984. DDT in Arctic burbot liver (Table 31) ranged from 4 to 219 ppb, while whitefish from the current samples ranged from only 0.2 to 3.6 ppb DDT. The previous data on whitefish are listed in Table 33 and they suggest somewhat lower levels of DDT in the current samples than was found in Cold Lake, Alberta, where up to 50 ppb was reported [157] and also lower than the value of 50 ppb in a round whitefish from Soldatna, Alaska [143]. Previous analyses of Arctic marine fish have indicated very low DDT levels, in the range of about 0.6 to 5 ppb [106]. Cod liver from the Gulf of St. Lawrence contained 1.1 ppm of toxaphene [110] and whitefish muscle from Lake Superior contained a range of toxaphene concentrations from 0.6 to 2.1 ppm [136].

The average daily intake of toxaphene in the U.S. has been estimated from the concentrations found in typical food items [45] and these results are listed in Table 30.

Table 30: Average U.S. daily intakes of toxaphene from various foods¹.

Food item	Toxaphene $\mu\text{g g}^{-1}$	Daily intake $\mu\text{g day}^{-1}$
Dairy products	0	0
Meat, fish and poultry	0.0007	0.192
Grain and cereal products	0.0023	0.979
Potatoes	0	0
Leafy vegetables	0.0015	0.0795
Legume vegetables	0	0
Root vegetables	0.0059	0.180
Garden fruits	0.0004	0.0268
Fruits	0	0
Oils and fats	0.0019	0.132
Sugar and adjuncts	0	0
Beverages	0	0
Total		1.59

¹From Gartrell *et al.* [45]

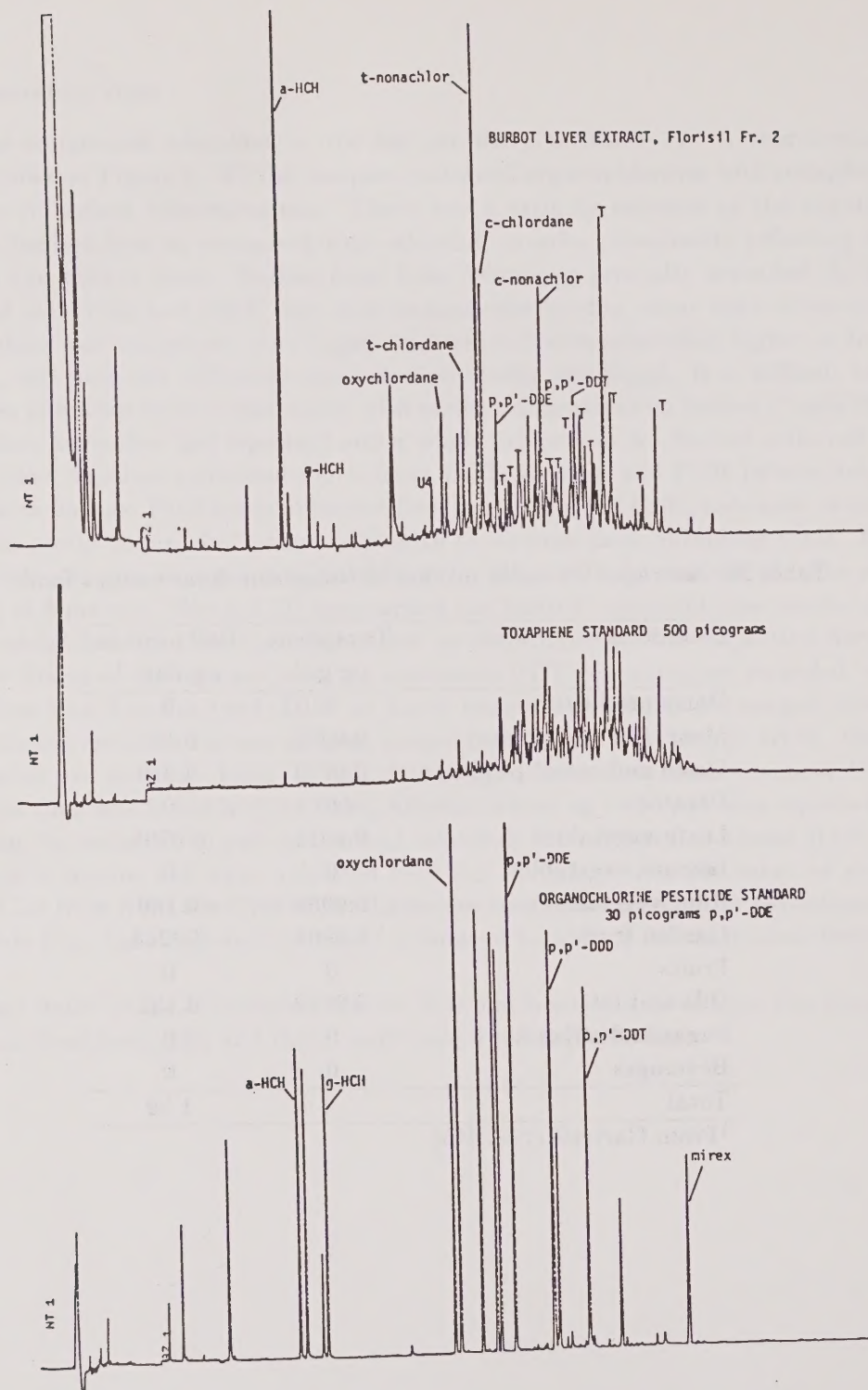


Figure 8: Representative gas chromatogram of organochlorine compounds (electron capture detector) extracted from burbot liver (top) with standards of toxaphene (middle) and other organochlorines (bottom).

Table 31: Organochlorines¹ (ng g⁻¹ wet weight) in arctic² burbot and whitefish, as compared with burbot from Lake Winnipeg

Source	CBz	HCH	DDT	Chlor- dane	Toxa- phene	Diel- drin	PCB tetra	PCB penta	PCB hexa	PCB hepta	PCB octa
Liver of burbot from arctic communities											
mean	17.1	13.9	43.2	83.1	815	5.9	20.5	36.3	49.9	27.0	5.0
S.D.	15.9	9.7	48.5	109.0	1241	4.8	26.4	31.2	42.2	21.3	6.4
min	1	4	4	5	27	nd	3	12	14	5	nd
max	71	36	219	241	5353	20	117	136	165	86	28
(n=18)											
Liver of burbot from Lake Winnipeg											
mean	9.1	14.7	259	38.5	387	9.8	116	249	359	240	60.4
S.D.	4.9	8.5	292	40.1	542	6.9	124	303	414	287	76.2
min	2	4	28	6	42	2	23	50	54	35	8
max	15	27	853	112	1655	21	347	922	1265	888	240
(n=8)											
Muscle of whitefish from arctic communities											
mean	.49	1.09	.95	1.79	25.6	0.95	1.56	3.04	2.74	1.37	0.18
S.D.	.50	.92	.98	2.52	26.2	3.19	1.68	1.57	1.76	1.06	0.21
min	nd	.1	.2	0.2	1.9	0.1	0.2	1.0	0.8	0.4	nd
max	1.8	3.8	3.5	9.8	85.8	13.3	5.6	6.7	7.4	4.0	0.7
n=17											

¹CBz - Sum of tetra-, penta-, and hexachlorobenzenes

HCH - Sum of α - β - and γ -hexachlorocyclohexanes

DDT - Sum of p,p'-DDT, p,p'-DDD, p,p'-DDE and o,p-DDT

Chlordane - Sum of chlordane-related peaks, calculated as chlordane

Toxaphene - calculated using 8 peaks in the GC profiles of toxaphene standard and of the samples

PCB (tetra) - Sum of tetrachloro-PCB congeners.

Sums of penta-, hexa-, hepta-, and octachloro- congeners are also shown.

²Arctic fish from Arctic Red River, Fort Franklin, Fort Simpson, Fort Good Hope, and Fort McPherson.

Table 32: Some published values for organochlorines in burbot

Source	Tissue	Contaminant	Range ($\mu\text{g g}^{-1}$)	Reference
Lake Superior 1977	whole fish	DDT	1.16–1.81	[4]
		PCB	1.40–1.59	
		Dieldrin	0.04–0.06	
		Chlordane	0.17–0.19	
		HCH	0.04–0.06	
Lake Superior 1977	whole fish	DDT	0.08–1.28	[162]
		PCB	0.5–1.7	
		Dieldrin	0.03	
Swiss waters	whole fish	PCB	0.1–5.6	[18]
Lake Geneva	whole fish	PCB	1.5	[104]
L. Paijanne, Finland	muscle	DDT	0.002–0.087	[50]
		PCB	0.013–0.224	
Fairbanks Alaska	whole fish	DDT	0.16	[143]
		PCB	0.22	
		Toxaphene	0.00	
		Chlordane	0.02	
		Dieldrin	0.00	
Churchill Falls Labrador	liver	PCB	0.04–1.11 ¹	[111]

¹Much higher values were reported in burbot from the tail race of the dam

The average U.S. dietary intake could be furnished by less than 1 gram of liver from some of the more highly contaminated northern burbot.

A recent report on toxaphene in fish from the Great Lakes [152] cited two earlier U.S. publications which bear on the question of the amount of toxaphene which can be consumed safely. In 1976 the U.S. Environmental Protection Agency estimated that the acceptable daily intake (a.d.i.) of toxaphene was 3.4 μg per kg of body weight per day. Then in 1977 the U.S. National Academy of Sciences gave the a.d.i. as only 1.25 μg per kg body weight per day. Thus a person weighing 70 kg might safely consume 87.5 μg per day using the lower estimate or 238 μg using the higher. The amount of liver required to furnish these amounts of toxaphene would be only about 17 and 45 grams in the worst case where calculation is based on the highest liver toxaphene concentration (5.35 ppm) found to date. It would clearly be desirable to know how much burbot liver is consumed by people in northern communities in order to determine whether it represents a hazardous source of toxaphene and other organochlorine compounds. Burbot liver has been reported to be the fish tissue most highly contaminated with organochlorines in Finland and Sweden, but, surprisingly, one recent study reported the absence of the pattern of chromatographic peaks typical of toxaphene [131].

Organochlorine compounds are widely distributed throughout the biosphere, including deep ocean sites [73] and animal life in Arctic and Antarctic regions where the compounds have not been used in quantity [125, 6, 175, 17, 115, 105]. These materials are transported from sources with movements of air [56, 35, 134] and water [48, 154]. Toxaphene has been reported in air from the Northwest Territories [10]. Hence it is not surprising to find the organochlorines present in Mackenzie River fish; rather it would be surprising to find them absent. Therefore, although it was not surprising to find toxaphene present in the Mackenzie River fish, finding concentrations comparable to southern fish was surprising. Several other organochlorines showed a distinct gradient of higher concentrations in the southern fish (Figure 9), but toxaphene had no such clear gradient (Figure 10).

If one assumes that the source of organochlorines is aerial dispersion from sources in lower latitudes of North/Central America, then the distribution of several organochlorines is what one would expect. With toxaphene, the distribution may indicate additional sources, such as some unknown use of toxaphene within the watershed, or perhaps aerial transport over the Arctic Ocean from Eurasia. A recent study of toxaphene components in air samples from Sweden suggested that the source might be eastern Europe [11]. Clearly these materials will require greater attention in future studies.

6.12 Metals

Concentration ranges for metals in burbot liver are given in Table 34, and those for whitefish muscle in Table 35, along with those few comparative data found. The major metals in Norman Wells oil are nickel and vanadium, and neither of these was found in whitefish muscle or burbot liver. The search for comparative data on metal content of fish was not extensive but those few data found were generally similar to the current results. Few other data on nickel or vanadium in these species have been published. Four whitefish from the Athabasca Delta were analyzed after a spill of refinery effluent into the Athabasca River and nickel levels ranged from 0.05 to 0.16 $\mu\text{g g}^{-1}$, with vanadium levels all below 0.1 $\mu\text{g g}^{-1}$ [Lockhart, unpublished]. As far as we are aware the metal concentrations would not be associated with pathology in the fish, however, some burbot livers at Fort Good Hope and Arctic Red River contained mercury at over 0.2 $\mu\text{g g}^{-1}$. The Medical Services Branch of Health and Welfare Canada has recommended 0.2 $\mu\text{g g}^{-1}$ as an upper safe limit for people who consume large quantities of fish [167, 3].

Table 33: Some published values for organochlorines in whitefish

Source	Tissue	Contaminant	Range ($\mu\text{g g}^{-1}$)	Reference
Lake Michigan (Lake whitefish)	muscle	PCB	1.7–4.4	[149]
Cold Lake area Alberta (Lake whitefish)	muscle	DDT	<0.0001–0.050	[157]
		PCB	nd–0.157	
		HCB	<0.0001–0.021	
		α HCH	<0.0001–0.042	
Lake Superior (Lake whitefish)	muscle	Toxaphene	0.6–2.1	[136]
Siskiwit Lake (Lake whitefish)	muscle?	Toxaphene	0.17–0.28	[127]
Soldatna, Alaska (Round whitefish)	whole fish	DDT	0.05	[143]
		PCB	0.0	
		Toxaphene	0.20	
		Chlordane	0.0	
		Dieldrin	0.0	
Tuktoyaktuk, NWT Broad whitefish	muscle	sDDT	0.0004	[106]
		sPCB	0.0018	
		α HCH	0.00003	
		γ HCH	<0.00005	
		HCB	0.00009	
		sChlordane	0.00048	
		Toxaphene	<0.002	
Inland lakes (Finland) Vendace (<i>Coregonus albula</i>)	muscle	sDDT	0.004–0.228	[132]
		sPCB	0.094–1.870	
		sChlordane	0.195–1.213	
	liver	sDDT	0.100–0.127	
		sPCB	0.941–1.668	
		sChlordane	0.069–0.715	

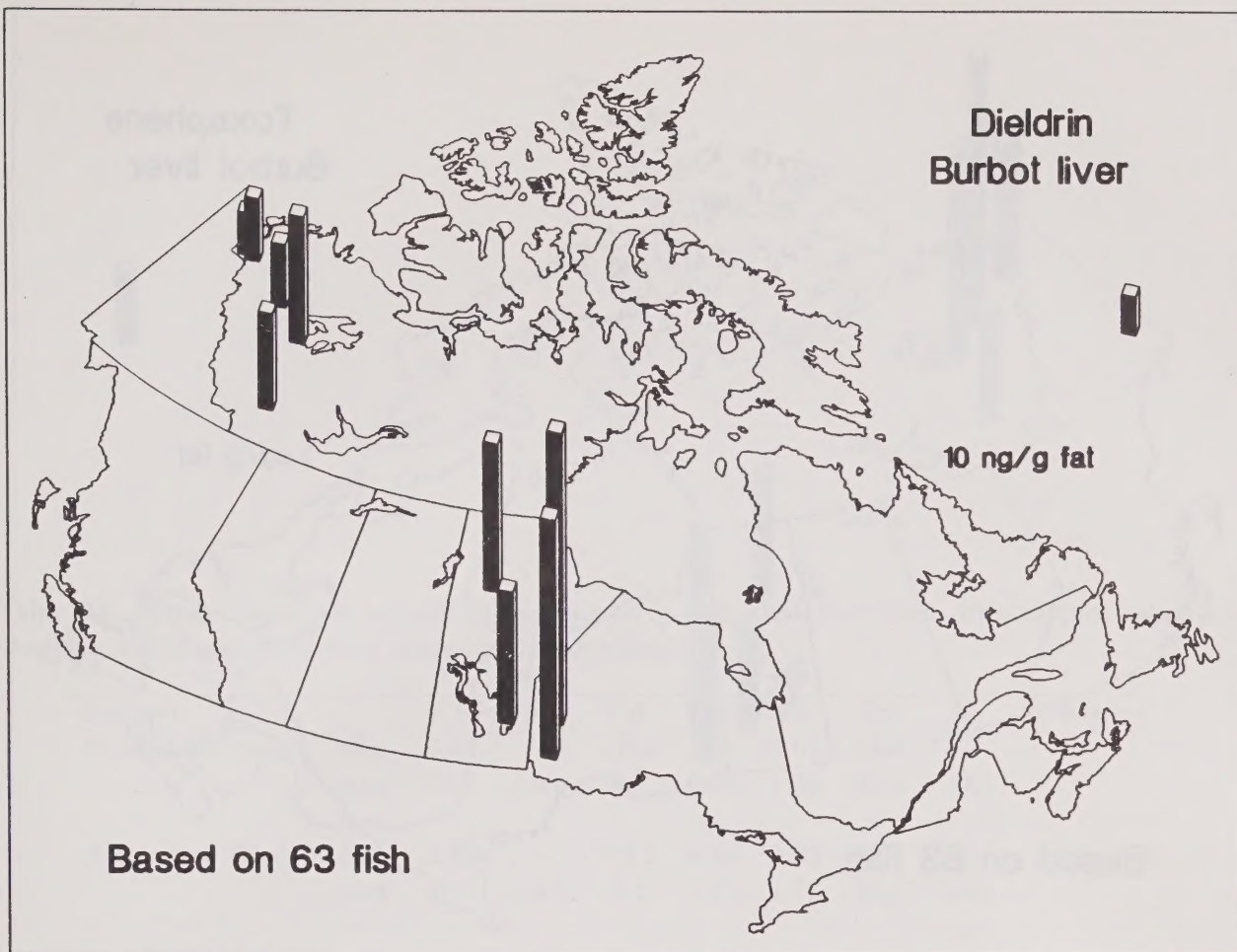


Figure 9: Map of mean dieldrin concentrations found in northern and southern burbot liver

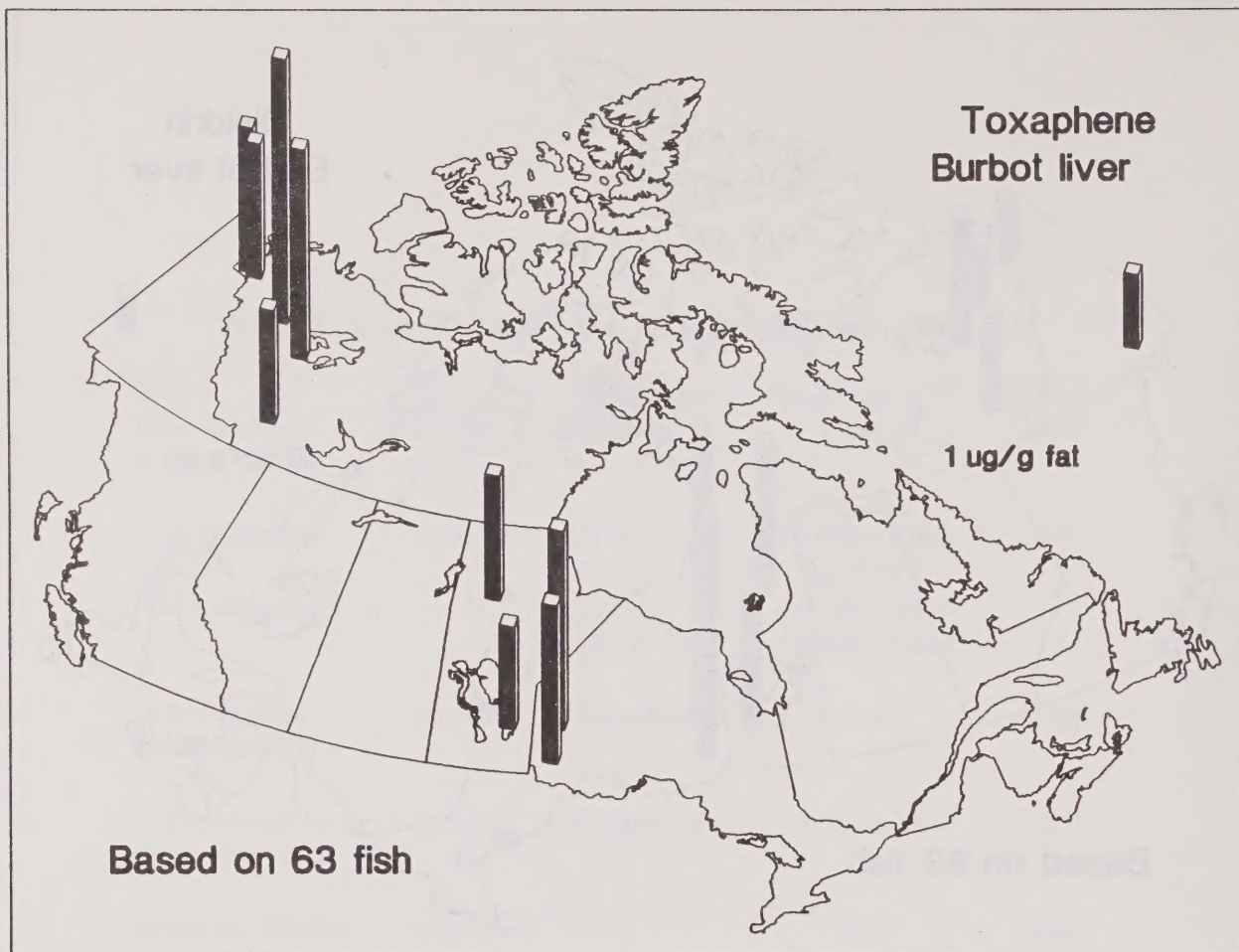


Figure 10: Map of mean toxaphene concentrations found in northern and southern burbot liver

Table 34: Ranges in metal concentrations ($\mu\text{g g}^{-1}$) in burbot liver from the samples taken in October, 1985, compared with other published values

Source		Cu	Ni	Cd	Hg	Se	Zn	V	Reference
Fort Good Hope	min	7.62	—	<0.1	0.06	0.81	14.3	—	
	max	17.7	<0.6	0.2	0.29	1.36	22.5	<0.1	
Arctic Red River	min	2.50	—	<0.1	0.05	0.35	12.4	—	
	max	7.84	<0.2	0.3	0.25	1.71	44.9	<0.1	
Fort Franklin	min	—	—	—	0.02	—	—	—	
	max	5.74	<0.5	<0.1	0.08	0.20	12.8	<0.1	
Fort Simpson	min	3.51	—	<0.1	0.03	0.81	12.8	—	
	max	22.3	<0.6	0.4	0.09	3.35	26.6	<0.1	
Fort Good Hope	min	1.60	—	<0.1	0.01	0.42	9.79	—	[165]
	max	30.1	—	0.42	0.25	2.87	79.3	—	
Lake Superior	mean	1.61	—	0.03	0.40	0.65	12.9	—	[4]
Whole burbot	mean	1.52	—	0.03	0.48	0.51	13.9	—	

Table 35: Ranges in metal concentrations ($\mu\text{g g}^{-1}$) in whitefish muscle from the samples taken in October, 1985, compared with other published values

Source		Cu	Ni	Cd	Hg	Se	Zn	V	Reference
Ft. Good Hope (Broad whitefish)	min	0.27	—	—	0.02	0.14	2.6	—	
	max	0.61	<0.2	<0.05	0.08	0.46	3.2	<0.1	
Arctic Red River (Lake whitefish)	min	0.24	—	—	0.04	0.20	2.2	—	
	max	0.62	<0.2	<0.05	0.14	0.59	2.9	<0.1	
Ft. Franklin (Lake whitefish)	min	0.19	—	—	0.08	0.15	2.7	—	
	max	0.30	<0.2	<0.05	0.13	0.29	3.5	<0.1	
Ft. Simpson (Lake whitefish)	min	0.17	—	—	0.02	0.27	2.8	—	
	max	0.56	<0.2	<0.05	0.18	1.03	6.3	<0.1	
Canadian Arctic (Whitefish spp.)	min	0.01	—	0.01	0.01	—	—	—	[172]
	max	1.10	—	0.10	0.04	—	—	—	
Tuktoyaktuk (Broad whitefish)	min	0.25	—	—	0.01	0.25	13.7	—	[106]
	max	0.70	—	<0.05	0.03	0.52	15.1	—	
Fort Good Hope (Lake whitefish)	min	0.07	—	—	0.06	0.28	3.01	—	[165]
	max	0.31	—	<0.01	0.23	0.62	4.12	—	
Lake Superior Whole bloater chub	mean	0.7	—	0.03	0.20	0.62	19.1	—	[4]
	mean	0.7	—	0.03	0.40	0.42	18.3	—	
Moose L. (Man.) L. Ontario (Lake whitefish)	mean	0.5	<0.2	<0.05	0.07	0.24	14.	—	[158]
	mean	0.94	<0.2	<0.05	0.17	0.38	12.	—	
Cold Lake, Alberta (Lake whitefish)	mean	0.51	—	—	0.16	—	6.22	3.72	[157]
Athabasca Delta (Lake whitefish)	min	—	0.05	—	—	—	—	—	[81]
	max	—	0.16	—	—	—	—	<0.1	
Lake Superior	mean	2.4	—	0.3	—	—	23.	—	[93]
Lake Michigan (Lake whitefish liver)	mean	8.5	—	0.09	—	—	—	—	

7 Conclusions

1. Dark coloured and small sized livers similar to those described by residents of Fort Good Hope and other northern communities were found to be prevalent in burbot from several northern communities both within and outside the Mackenzie River drainage. The appearance of the liver is linked to the energy stored in the liver, and the condition can be produced experimentally by reducing liver fats. The cause of the liver condition is unlikely to be exposure to petroleum hydrocarbons, since it occurs remote from petroleum sources, and since it can be induced by starving the fish. However, petroleum or other pollution cannot be ruled out conclusively. If the cause of the condition of burbot liver does include exposure to petroleum hydrocarbons, then clearly Norman Wells is not the exclusive source since the condition was as prevalent at Fort Franklin as at Fort Good Hope or Arctic Red River.
2. The most specific diagnostic test available, namely liver mixed-function oxidase enzymes, indicated that the northern burbot were not responding to oil pollution. However, this interpretation may have been complicated by the observation that the 'control' population of burbot from Lake Winnipeg had higher levels of PCBs than the arctic burbot, and this could have elevated the controls and hence masked a very low level oil induction in the arctic fish.
3. The fish from all locations sampled were contaminated with low levels of low-boiling aromatic hydrocarbons typically present in Norman Wells crude oil and in refined petroleum products. The extent of this contamination indicated widespread occurrence of these compounds, however, the level of this contamination at Fort Good Hope was several times higher in late winter than in late fall, which suggests an additional source of the hydrocarbons under river ice. The winter *vs.* summer difference was not evident at Fort Simpson, which further suggests that an under-ice source might be located between Fort Simpson and Fort Good Hope.
4. There are only anecdotal reports on the timing of the appearance of the small, dark livers in burbot. If it has indeed become prevalent only recently, then it is legitimate to search for recent changes in the environment that might offer some explanation. While sample sizes are very small, the limited data do not indicate a decrease in the growth rates of the fish taken in 1985 as compared with those in the early 1970s. Clearly the liver condition is related to the liver content of fat and calories, and such a relationship suggests the likelihood of a linkage to growth. The absence of a change in growth rate then suggests that the liver condition may have been present but not reported for some time. Earlier Alaskan studies suggested a linkage between small livers and maturation for spawning, with fish having small livers being unlikely to spawn due to low energy storage. If this hypothesis holds, then the anecdotal recent increases in the abundance of small livers suggests that a smaller proportion of the burbot may be spawning than was the case historically.
5. There is widespread contamination of fish from the Mackenzie River basin with several organochlorine compounds, notably toxaphene, chlordane, and PCBs. Future study will be needed to define the geographic extent of this contamination, trends in contaminant levels over time, and the implications of the contamination for arctic ecosystems and for consumers of products from those ecosystems.
6. The complaint regarding 'watery' flesh of whitefish is also borne out by the analyses. The northern whitefish have higher water content and lower fat content than most other samples of whitefish for which data have been found, but investigation of this matter remains incomplete.

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References

- [1] R.F. Addison, M.E. Zinck, and D.E. Willis. Mixed function oxidase systems in trout (*Salvelinus fontinalis*) liver: Absence of induction following feeding of *p,p'*-DDT or *p,p*-DDE. *Comp. Biochem. Physiol.*, 57C:39-43, 1977.
- [2] Anonymous. *Polynuclear aromatic hydrocarbons - A background report including available Ontario data*, volume 58-79 of *ARB-TDA-Report*. Ontario Ministry of the Environment, 1979. 193 pg.
- [3] Anonymous. Methylmercury in Canada. Exposure of Indian and Inuit residents to methylmercury in the Canadian environment. Report 2, Health and Welfare Canada, Medical Services Branch, 1984.
- [4] F.A.J. Armstrong and A. Lutz. Lake Superior, 1974: Chlorinated insecticides, heavy metals, and radioactivity in offshore fish. Technical Report of Fisheries and Marine Service 683, Department of the Environment, 1977. 17 pg.
- [5] F.A.J. Armstrong and J.F. Uthe. Semi-automated determination of mercury in animal tissue. *Atomic Absorption Newsletter*, 10:101-103, 1972.
- [6] K. Ballschmiter, Ch. Scholz, H. Buchert, M. Zell, K. Figge, and K. Polzhofer. Studies of the global baseline pollution. V. Monitoring the baseline pollution of the Sub-Antarctic by penguins as bioindicators. *Fresenius Z. Anal. Chem.*, 309:1-7, 1981.
- [7] P.C. Baumann, W.D. Smith, and M. Ribick. Hepatic tumor rates and polyaromatic hydrocarbon levels in two populations of brown bullhead (*Ictalurus nebulosus*). In M.W. Cooke, A.J. Dennis, and G.L. Fisher, editors, *Polynuclear Aromatic Hydrocarbons: Sixth International Symposium on Physical and Biological Chemistry*, pages 93-102, Columbus, Ohio, 1982. Battelle Press.
- [8] N. Berg. Chemical and sensory analysis of off-flavours in fish from polluted rivers in Norway. *Water Sci. Technol.*, 16:59-64, 1983.
- [9] L.M.J. Bernier. Liver atrophy of burbot, *Lota lota* (L.), infected with larvae of the nematode *Raphidascaris acus* (Bloch, 1779) and with plerocercoids of the cestode *Triaenophorus nodulosus* (Pallas, 1760), with notes on the presence of *R. acus* in fishes of the family *Corigonidae*. Unpublished Report, 36 pg, 1986.
- [10] T.F. Bidleman, C.P. Rice, and C.E. Olney. High molecular weight chlorinated hydrocarbons in the air and sea: Rates and mechanisms of air/sea transfer. In H.L. Windom and R.A. Duce, editors, *Marine Pollutant Transfer*, pages 323-351. Lexington Books, Lexington, Mass., and Toronto. D.C. Heath and Co., 1976.
- [11] T.F. Bidleman, U. Wideqvist, B. Jansson, and R. Soderlund. Organochlorine pesticides and polychlorinated biphenyls in the atmosphere of southern Sweden. *Atmos. Environ.*, 21:641-654, 1987.
- [12] E.C. Black, N.J. Bosomworth, and G.E. Docherty. Combined effect of starvation and severe exercise on glycogen metabolism of rainbow trout, *Salmo gairdneri*. *J. Fish. Res. Bd. Canada*, 23:1461-1463, 1966.

- [13] J.J. Black. Field and laboratory studies on environmental carcinogenesis in Niagara River fish. *J. Great Lakes Res.*, 9:326-334, 1983.
- [14] R.G. Bligh and W.J. Dyer. A rapid method of total lipid extraction and purification. *Can. J. Biochem. Physiol.*, 37:911-917, 1959.
- [15] A.M. Bobra, W.Y. Shiu, and D. Mackay. A predictive correlation for the acute toxicity of hydrocarbons and chlorinated hydrocarbons to the water flea (*Daphnia magna*). *Chemosphere*, 12:1121-1129, 1983.
- [16] R.M. Bone and R.J. Mahnic. Norman Wells: the oil center of the Northwest Territories. *Arctic*, 37:53-60, 1984.
- [17] G.W. Bowes and C.J. Jonkel. Presence and distribution of polychlorinated biphenyls (PCB) in arctic and subarctic marine food chains. *J. Fish. Res. Bd. Canada*, 32:2111-2123, 1975.
- [18] G. Burgermeister, M. Bedrani, and J. Tarradellas. Utilisation de la lotte comme indicateur de la pollution des eaux continentales par des polluants organochlores. *Eau du Québec*, 16:135-143, 1983.
- [19] M.D. Burke and R.T. Mayer. Ethoxyresorufin: Direct fluorometric assay of microsomal O-dealkylation which is preferentially induced by 3-methylcholanthrene. *Drug. Metab. Disp.*, 2:583-588, 1974.
- [20] I.E. Bykhovskaya-Pavlovskaya, A.V. Gusev, M.N. Dubinina, N.A. Izyumova, T.S. Smirnova, I.L. Sokolovskaya, G.A. Shtein, S.S. Shul'man, and V.M. Epshtein. *Key to Parasites of Freshwater Fish of the U.S.S.R.*, volume 80. Academy of Sciences of the USSR, 1962. Edited by D.E. Byknovskii, Translated by A. Birron and Z.S. Cole. Israel Program for Scientific Translations, Jerusalem, 1964.
- [21] L-C. Chen. The biology and taxonomy of the burbot, *Lota lota leptura*, in interior Alaska. *Biol. Pap. Univ. Alaska*, 11:53 pg, 1969.
- [22] J. Cochin and J. Axelrod. Biochemical and pharmacological changes in rat following chronic administration of morphine, nalorphine, and normorphine. *J. Pharmacol. Exper. Therap.*, 125:105-110, 1959.
- [23] J.F. Craig. The body composition of adult perch, *Perca fluviatilis* in Windemere, with reference to seasonal changes and reproduction. *J. Anim. Ecol.*, 46:612-632, 1977.
- [24] J.F. Craig, M.J. Kenley, and J.F. Talling. Comparative estimations of the energy content of fish tissue from bomb calorimetry, wet oxidation and proximate analysis. *Freshwater Biol.*, 8:585-590, 1978.
- [25] J.M. Daisey, R.J. McCaffrey, and R.A. Gallagher. Polycyclic aromatic hydrocarbons and total extractable particulate organic matter in the Arctic aerosol. *Atmos. Environ.*, 15:1353-1363, 1981.
- [26] J.M. Davies, J.S. Bell, and C. Houghton. A comparison of the levels of hepatic aryl hydrocarbon hydroxylase in fish caught close to and distant from North Sea oil fields. *Mar. Environ. Res.*, 14:23-45, 1984.

- [27] E. deHullu and C.L.M. Poels. Internal Report, KIWA. (cited from Helder (1980), original paper not seen), 1976.
- [28] J.W. Depierre, M.S. Moron, K.A.M. Johannesen, and L. Ernster. A reliable, sensitive and convenient radioactive assay for benzpyrene monooxygenase. *Anal. Biochem.*, 63:470-484, 1975.
- [29] A.C. Dey, J.W. Kiceniuk, U.P. Williams, R.A. Khan, and J.F. Payne. Long term exposure of marine fish to crude petroleum - I, studies on liver lipids and fatty acids in cod (*Gadus morhua*) and winter flounder (*Pseudopleuronectes americanus*). *Comp. Biochem. Physiol.*, 75C:93-101, 1983.
- [30] R.L. Dixon, R.W. Shultice, and J.R. Fouts. Factors affecting drug metabolism by liver microsomes. IV. Starvation. *Proc. Soc. Exp. Biol. Med.*, 103:333-335, 1960.
- [31] M. Dubois, K.A. Gilles, J.K. Hamilton, P.A. Rebers, and F. Smith. Colorimetric method for determination of sugars and related substances. *Anal. Chem.*, 28:350-356, 1956.
- [32] L.C. Dugal. Proximate composition of some freshwater fish. Tech. Circ. 5, Fish. Res. Bd. Canada, London Biol. Stn., 1962. 6 pg.
- [33] D.A. Duncan. Sublethal and chronic effects of oil/dispersant mixtures on larval rainbow trout (*Salmo gairdneri*). Freshwater oil spill research program. freshwater institute component. final report for 1983/84 research activity, Report to Department of Fisheries and Oceans, 1984. 44-84.
- [34] D.A. Duncan. Effects of oil and oil/dispersant mixtures on survival, edema development, growth, tissue structure, osmoregulation and enzyme activity. Freshwater oil spill research program. freshwater institute component. final report for 1984/85 research activity, Report to Department of Fisheries and Oceans, 1985. 24 pg + apps.
- [35] S.J. Eisenreich, G.J. Hollod, and T.C. Johnson. Accumulation of polychlorinated biphenyls (PCBs) in surficial Lake Superior sediments. Atmospheric deposition. *Environ. Sci. Technol.*, 13:569-573, 1979.
- [36] B.J.M. Elliott. Body composition of brown trout (*Salmo trutta* L.) in relation to temperature and ration size. *J. Animal Ecol.*, 45:273-289, 1976.
- [37] G.W. Esch, J.W. Gibbons, and J.E. Borque. An analysis of the relationship between stress and parasitism. *Amer. Midland Nat.*, 93:339-353, 1975.
- [38] R.W. Estabrooke, J. Peterson, J. Baron, and A. Hildebrandt. The spectrophotometric measurement of turbid suspensions of cytochromes associated with drug metabolism. In C.R. Chignell, editor, *Methods in Pharmacology 2*, pages 303-350. Appleton-Century-Crofts, New York, 1972.
- [39] F.W. Fales. Evaluation of a spectrophotometric method for for determination of total fecal lipid. *Clin. Chem.*, 17:1103-1108, 1971.
- [40] T. Fazio and J.W. Howard. Polycyclic aromatic hydrocarbons in foods. In A. Bjorseth, editor, *Handbook of Polycyclic Aromatic Hydrocarbons*, pages 461-505. Marcel Dekker, Inc., New York, 1983.

- [41] L. Forlin. Effects of Clophen A50, 3-methylcholanthrene, pregnenolone-16 α -carbonitrile, and phenobarbital on the hepatic microsomal cytochrome P-450 dependent monooxygenase system in rainbow trout, *Salmo gairdneri* of different age and sex. *Toxicol. Appl. Pharmacol.*, 54:420-430, 1980.
- [42] L. Forlin and U. Lidman. O-demethylation of p-nitroanisole by mixed function oxidase system of the rainbow trout (*Salmo gairdneri*) liver. *Gen. Pharmacol.*, 10:411-415, 1979.
- [43] C.R. Fremling. Impacts of a spill of No 6 fuel oil on Lake Winona. In *Proc. 1981 Oil Spill Conf.*, pages 419-421, Washington, 1981. Amer. Petrol. Inst.
- [44] R.B. Gammage. Polycyclic aromatic hydrocarbons in work atmospheres. In A. Bjorseth, editor, *Handbook of Polycyclic Aromatic Hydrocarbons*, pages 653-707. Marcel Dekker Inc., New York, 1983.
- [45] M.J. Gartrell, J.C. Craun, D.S. Podrebarac, and E.L. Gunderson. Pesticides, selected elements, and other chemicals in adult total diet samples, October 1980-March 1982. *J. Assoc. Off. Anal. Chem.*, 69:146-161, 1986.
- [46] K. Grob and G. Grob. Organic substances in potable water and its precursor. Part II. Applications in the area of Zurich. *J. Chromatog.*, 90:303-313, 1974.
- [47] W.E. Haensly, J.M. Neff, J.R. Sharp, A.C. Morris, M.F. Bedgood, and P.D. Boem. Histopathology of *Pleuronectes platessa* L. from Aber Wrac'h and Aber Benoit, Brittany, France: Long-term effects of the Amoco Cadiz crude oil spill. *J. Fish Diseases*, 5:365-391, 1982.
- [48] G.R. Harvey and W.G. Steinhauer. Transport pathways of polychlorinated biphenyls in Atlantic water. *J. Mar. Res.*, 34:561-575, 1976.
- [49] R.M. Haslemore and P.G. Roughan. Rapid chemical analysis of some plant constituents. *J. Sci. Food Agric.*, 27:1171-1178, 1976.
- [50] M.L. Hattula, J. Janatuinen, J. Sarkka, and J. Passivirta. A five-year monitoring study of the chlorinated hydrocarbons in the fish of a Finnish lake ecosystem. *Environ. Pollut.*, 15:121-139, 1978.
- [51] J.W. Hawkes. The effects of petroleum hydrocarbon exposure on the structure of fish tissues. In D.A. Wolfe, editor, *Fate and Effects of Petroleum Hydrocarbons in Marine Organisms and Ecosystems*, pages 115-128. Pergamon, 1977.
- [52] J.R. Hazel. The influence of temperature adaptation on the composition of the neutral lipid fraction of rainbow trout (*Salmo gairdneri*) liver. *J. Exp. Zool.*, 207:33-42, 1979.
- [53] T. Helder. Effects of 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD) on early life stages of the pike (*Esox lucius*). *Sci. Tot. Environ.*, 14:255-264, 1980.
- [54] R.J. Henry. *Clinical Chemistry*. Harper and Row, 1964.
- [55] L.C. Hewson. Age, maturity, spawning and food of burbot *Lota lota leptura*, in Lake Winnipeg. *J. Fish. Res. Bd. Canada*, 12:930-940, 1955.
- [56] R.M. Hoff and K.-W. Chan. Atmospheric concentrations of chlordane at Mould Bay, N.W.T., Canada. *Chemosphere*, 15:449-452, 1986.

- [57] D.G. Iredale and R.K. York. *A guide to handling and preparing freshwater fish*. Department of Fisheries and Oceans, Freshwater Institute, Winnipeg, 1985. 47 pg.
- [58] M.O. James and J.R. Bend. Polycyclic aromatic hydrocarbon induction of cytochrome P-450 mixed-function oxidases in marine fish. *Toxicol. Appl. Pharmacol.*, 54:117-133, 1980.
- [59] M.O. James and P.J. Little. Polyhalogenated biphenyls and phenobarbital: Evaluation as inducers of drug metabolizing enzymes in the sheepshead, *Archosargus probatocephalus*. *Chem.-Biol Interactions*, 36:229-248, 1981.
- [60] P.M. Jangaard, R.G. Ackman, and J.C. Sipos. Seasonal changes in fatty acid composition of cod liver, flesh, roe, and milt lipids. *J. Fish. Res. Bd. Canada*, 24:613-627, 1967.
- [61] P.M. Jangaard, H. Brockerhoff, R.D. Burgher, and R.J. Hoyle. Seasonal changes in general condition and lipid content of cod from inshore waters. *J. Fish. Res. Bd. Canada*, 24:607-612, 1967.
- [62] K.A.M. Johannesen and J.W. DePierre. Measurement of cytochrome P-450 in the presence of large amounts of contaminating hemoglobin and methemoglobin. *Anal. Biochem.*, 86:725-732, 1978.
- [63] D.B. Josephson, R.C. Lindsay, and D.B. Stuibier. Identification of compounds characterizing the aroma of fresh whitefish. *J. Agric. Food Chem.*, 31:326-330, 1983.
- [64] D.B. Josephson, R.C. Lindsay, and D.B. Stuibier. Variations in the occurrences of enzymatically derived volatile aroma compounds in salt- and freshwater fish. *J. Agric. Food Chem.*, 32:1344-1347, 1984a.
- [65] D.B. Josephson, R.C. Lindsay, and D.B. Stuibier. Biogenesis of lipid-derived volatile aroma compounds in the emerald shiner (*Notropis atherinoides*). *J. Agric. Food Chem.*, 32:1347-1352, 1984b.
- [66] D.B. Josephson, R.C. Lindsay, and D.B. Stuibier. Identification of volatile aroma compounds from oxidized frozen whitefish (*Coregonus clupeaformis*). *Can. Inst. Food Sci. Technol. J.*, 17:178-182, 1984c.
- [67] F. Juttner. Detection of lipid degradation products in the water of a reservoir during a bloom of *Synura uvella*. *Appl. Environ. Microbiol.*, 41:100-106, 1981.
- [68] F. Juttner. Analysis of organic compounds (VOC) in the forest air of the southern black forest. *Chemosphere*, 15:985-992, 1986.
- [69] R. Kato and J.R. Gillette. Effect of starvation on NADPH dependent enzymes in liver microsomes of male and female rats. *J. Pharm. Exp. Therap.*, 150:279-284, 1965.
- [70] F. Kh. Khalilov. Some material on the histology and histochemistry of the pancreas and liver of teleost fishes. *J. Ichthyol.*, 8:246-249, 1968.
- [71] G. Kinzer, R. Riggan, T. Bishop, M. Birts, and P. Strup. EPA method study 20: Method 610 - PNAs. Report EPA-600/4-84-063., U.S. Environmental Protection Agency, 1984.
- [72] M. Kleiber. *The Fire of Life - An Introduction to Animal Energetics*. R.E. Kreiger Publishing, Huntington, NY, 1975.

- [73] A.H. Knap, K.S. Binkley, and W.G. Deuser. Synthetic organic chemicals in the deep Sargasso Sea. *Nature*, 319:572-574, 1986.
- [74] S. Korn, J.W. Strushaker, and P. Benville Jr. Effects of benzene on growth, fat content, and caloric content of striped bass, *Morone saxatilis*. *Fishery Bull.*, 74:694-698, 1976.
- [75] M.M. Krahn, L.D. Rhodes, M.S. Myers, L.K. Moore, W.D. MacLeod Jr., and D.C. Malins. Associations between metabolites of aromatic compounds in bile and the occurrence of hepatic lesions in English sole (*Papophrys vetulus*) from Puget Sound, Washington. *Arch. Environ. Contam. Toxicol.*, 15:61-67, 1986.
- [76] A. Larsson and K. Lewander. Metabolic effects of starvation in the eel, *Anguilla anguilla* L. *Comp. Biochem. Physiol.*, 44A:367-374, 1973.
- [77] J.F. Lawrence and D.F. Weber. Determination of polycyclic aromatic hydrocarbons in some Canadian commercial fish and shellfish, and meat products by liquid chromatography with confirmation by capillary gas chromatography-mass spectrometry. *J. Agric. Food Chem.*, 32:789-794, 1984.
- [78] J.J. Lech, M.J. Vodicknik, and C.R. Elcombe. Induction of monooxygenase activity in fish. In L.J. Weber, editor, *Aquatic Toxicology*, pages 107-148, New York, 1982. Raven Press.
- [79] R.F. Lee, R. Sauerheber, and G.H. Dobbs. Uptake, metabolism and discharge of polycyclic aromatic hydrocarbons by marine fish. *Mar. Biol.*, 17:201-208, 1972.
- [80] W.L. Lockhart. Methoxychlor studies with fish: Athabasca River exposures and experimental exposures. In W.O. Haufe and G.C.R. Croome, editors, *Control of Black Flies in the Athabasca River*, pages 183-196. Alberta Environment, Tech. Rep. Poll. Control Div., Edmonton, 1980.
- [81] W.L. Lockhart, 1982. Unpublished data.
- [82] W.L. Lockhart, R.W. Danell, and D.A.J. Murray. Acute toxicity bio-assays with petroleum: Influence of exposure conditions. In S. Hrudy and J. Vandermeulen, editors, *Oil and Freshwater: Chemistry, Biology and Countermeasure Technology*, pages 335-344. Pergamon Press, 1986.
- [83] W.L. Lockhart, R.W. Danell, D.A.J. Murray, R.K. York, D. Tilden, and K. Hall. Retention of deleterious properties by diesel fuel spilled in northern Canada in winter. Manuscript in preparation, 1986.
- [84] W.L. Lockhart, D.A. Metner, and D.C.G. Muir. Analysis of liver mixed-function oxidase enzymes of harp seals and induction of these enzymes by petroleum. Report to Panel on Energy Research and Development, In preparation, 1989.
- [85] W.L. Lockhart, D.A. Metner, D.A.J. Murray, and D.C.G. Muir. Hydrocarbons and complaints about fish quality in the Mackenzie River, Northwest Territories, Canada. *Water Poll. Res. J. Canada*, 22:616-628, 1987.
- [86] W.L. Lockhart and D.C.G. Muir. Trace organic contaminants in the Canadian Arctic. Proceedings 40th annual meeting, Western Canada Water and Wastewater Assoc., 1988. In Press.

- [87] W.L. Lockhart, R.K. York, and R.W. Danell. Tainting of trout and charr fillets by petroleum oil. Manuscript in preparation, 1989.
- [88] R.M. Love. Studies on the North Sea cod III. Effects of starvation. *Jour. Sci. Food Agric.*, 9:617-620, 1958.
- [89] R.M. Love. *The Chemical Biology of Fishes*. Academic Press, London, first edition, 1970. 547 pg.
- [90] R.M. Love. *The Chemical Biology of Fishes*, volume 2. Academic Press, London, 1980. 943 pg.
- [91] O.H. Lowry, N.J. Rosebrough, A.L. Farr, and R.J. Randall. Protein measurement with the Folin phenol reagent. *J. Biol. Chem.*, 193:265-275, 1951.
- [92] A.H.Y. Lu, A. Somogyi, S. West, R. Kuntzman, and A.H. Conney. Pregnenolone-16a-carbonitrile: a new type of inducer for drug metabolizing enzymes. *Arch. Biochem. Biophys.*, 152:457-462, 1972.
- [93] H.F. Jr. Lucas, D.N. Edgington, and P.J. Colby. Concentrations of trace elements in Great Lakes fishes. *J. Fish. Res. Board Canada*, 27:677-684, 1970.
- [94] P.L. Luxon, P.V. Hodson, and U. Borgmann. Hepatic aryl hydrocarbon hydroxylase activity of lake trout (*Salvelinus namaycush*) as an indicator of organic pollution. *Environ. Toxicol. Chem.*, 6:649-657, 1987.
- [95] A.E. Maccubbin, P. Black, L. Trzeciak, and J.J. Black. Evidence for polynuclear aromatic hydrocarbons in the diet of bottom-feeding fish. *Bull. Environ. Contam. Toxicol.*, 34:876-882, 1985.
- [96] J. Mai, J.K. Shetty, T-M. Kan, and J.E. Kinsella. Protein and amino acid composition of select freshwater fish. *J. Agric. Food Chem.*, 28:884-885, 1980.
- [97] M.A.K. Markwell, S.M. Haas, N.E. Tolbert, and L.L. Bieber. Protein determination in membrane and lipoprotein samples: manual and automated procedures. *Methods Enzymol.*, 72:296-303, 1981.
- [98] F.L. Mayer, P.M. Mehrle, and P.L. Crutcher. Interactions of toxaphene and vitamin C in channel catfish. *Trans. Amer. Fish. Soc.*, 107:326-333, 1978.
- [99] P. Mazel. General principles and procedures for drug metabolism *in vitro*. In G. Mandel B.N LaDu and E.L. Way, editors, *Fundamentals of Drug Metabolism and Disposition*, pages 546-582. Kreiger Publishing Co., Malabar, Florida, 1971.
- [100] N.D. Mazmanidi and G.I. Kovaleva. The effect of dissolved petroleum products on some of the aspects of carbohydrate metabolism in fishes and invertebrates. *J. Ichthyol.*, 15:811-816, 1975.
- [101] C.D. Minchew and J.D. Yarbrough. The occurrence of fin rot in mullet (*Mugil cephalus*) associated with crude oil contamination of an estuarine pond-ecosystem. *J. Fish Biol.*, 10:319-323, 1977.
- [102] R. Montgomery. Determination of glycogen. *Arch. Biochem. Biophys.*, 67:378-386, 1957.

- [103] T. Motohiro. Tainted fish caused by petroleum compounds - a review. *Water Sci. Tech.*, 15 (Finland):75-83, 1983.
- [104] J. Mowrer, K. Aswald, G. Burgermeister, L. Machado, and J. Tarradellas. PCB in a Lake Geneva ecosystem. *Ambio*, 11:355-358, 1982.
- [105] D.C.G. Muir, R.J. Norstrom, and M. Simon. Organochlorine contaminants in arctic marine food chains: Accumulation of specific polychlorinated biphenyls and chlordane-related compounds. *Environ. Sci. Technol.*, 22:1071-1079, 1988.
- [106] D.C.G. Muir, R. Wagemann, W.L. Lockhart, N.P. Grift, B. Billeck, and D. Metner. Heavy metal and organic contaminants in arctic marine fishes. Technical Report Environmental Studies No. 42, Department of Indian Affairs and Northern Development, Northern Affairs Program, Ottawa, 1986. NOGAP Project A-12. 64 pg.
- [107] R.A. Murchelano and R.E. Wolke. Epizootic carcinoma in the winter flounder *Pseudopleuronectes americanus*. *Science*, 228:587-589, 1983.
- [108] D.A.J. Murray and W.L. Lockhart. Determination of trace volatile organic compounds in fish tissues by gas chromatography. *J. Assoc. Off. Anal. Chem.*, 71:1086-1089, 1988.
- [109] D.A.J. Murray, W.L. Lockhart, and G.R.B. Webster. Analysis of the water-soluble fraction of crude oils and petroleum products by gas chromatography. *Oil Petrochem. Poll.*, 2:39-46, 1984.
- [110] C.J. Musial and J.F. Uthe. Widespread occurrence of the pesticide toxaphene in Canadian east coast marine fish. *Intern. J. Environ. Anal. Chem.*, 14:117-126, 1983.
- [111] C.J. Musial, J.F. Uthe, R.J. Wiseman, and R.A. Matheson. Occurrence of PCB residues in burbot (*Lota lota*) and lake trout (*Salvelinus manaycush*) from the Churchill Falls power development area. *Bull. Environ. Contam. Toxicol.*, 23:256-261, 1979.
- [112] T. Nash. The colorimetric estimation of formaldehyde by means of the Hantzsch reaction. *Biochemistry*, 55:416-421, 1953.
- [113] J.M. Neff. *Polycyclic Aromatic Hydrocarbons in the Aquatic Environment. Sources, Fates and Biological Effects*. Applied Science Publishers, London, 1979. 262 pg.
- [114] A.J. Niimi and V. Palazzo. Biological half-lives of eight polycyclic aromatic hydrocarbons (PAHs) in rainbow trout (*Salmo gairdneri*). *Water Res.*, 20:503-507, 1986.
- [115] R.J. Norstrom and D.C.G. Muir. Long-range transport of organochlorines in the Arctic and sub-Arctic: evidence from the analysis of marine mammals and fish. In N.W. Schmidke, editor, *Toxic Contaminants in the Great Lakes. I. Chronic Effects of Toxic Contaminants in Large Lakes*, pages 83-112. Lewis Publishing Co., Chelsea, Michigan, 1986.
- [116] R.J. Norstrom and H.T. Won. Long-term preservation of egg and tissue homogenates for determination of organochlorine compounds: freezing vs. freeze-drying. *J. Assoc. Offic. Anal. Chem.*, 68:130-135, 1985.
- [117] M. Ogata and Y. Miyake. Offensive-odor substance in the evil-smelling fish from the sea polluted by petroleum and petrochemical industrial waste. Report 1. identification of offensive-odor substance. *Acta Med. Okayama*, 24:471-481, 1970.

- [118] M. Ogata and Y. Miyake. Identification of substances in petroleum causing objectionable odour in fish. *Water Res.*, 7:1493-1504, 1973.
- [119] T. Omura and R. Sato. The carbon monoxide-binding pigment of liver microsomes I. Evidence for its hemoprotein nature. *J. Biol. Chem.*, 239:2370-2378, 1964a.
- [120] T. Omura and R. Sato. The carbon monoxide-binding pigment of liver microsomes II. Solubilization, purification, and properties. *J. Biol. Chem.*, 239:2379-2385, 1964b.
- [121] J.F. Payne. Field evaluation of benzopyrene hydroxylase induction as a monitor for marine petroleum pollution. *Science*, 191:945-946, 1976.
- [122] J.F. Payne, L.L. Fancey, A.D. Rahimtula, and E.L. Porter. Review and perspective on the use of mixed-function oxygenase enzymes in biological monitoring. *Comp. Biochem. Physiol.*, 86C:233-245, 1987.
- [123] Jr. P.E. Benville and S. Korn. The acute toxicity of six monocyclic aromatic crude oil components to striped bass (*Monone saxatilis*) and bay shrimp (*Crago franciscorum*). *Calif. Fish and Game*, 63:204-209, 1977.
- [124] M.G. Pedersen, W.K. Hershberger, P.K. Zachariah, and M.R. Juchau. Hepatic biotransformation of environmental xenobiotics in six strains of rainbow trout (*Salmo gairdneri*). *J. Fish. Res. Board Canada*, 33:666-675, 1976.
- [125] T.J. Peterle. DDT in Antarctic snow. *Nature*, 224:620, 1969.
- [126] G.K. Petrushevski and S.S. Shulman. The parasitic diseases of fishes in the natural waters of the USSR. In G.K. Petrushevski V. Dogiel and Yu.I. Polanski, editors, *Parasitology of Fishes* (translated by Z. Kabata), pages 299-334. Oliver and Boyd, Edinburgh and London, 1961.
- [127] J.D. Petty, T.R. Schwartz, and D.L. Stalling. Gas chromatographic residue patterns of toxaphene in fish samples from the Great Lakes and from rivers of the southeastern United States. In T.P. Boyle, editor, *New Approaches to Monitoring Aquatic Ecosystems*, pages 165-172. American Society for Testing and Materials, Philadelphia, 1987. ASTM STP 940.
- [128] R.J. Pohl and J.R. Fouts. A rapid method for assaying the metabolism of 7-ethoxyresorufin by microsomal cellular fractions. *Anal. Biochem.*, 107:150-155, 1980.
- [129] R.A. Prough, M.D. Burke, and R.T. Mayer. Direct fluorometric methods for measuring mixed function oxidase activity. *Methods Enzymol.*, LII (C):372-377, 1978.
- [130] R.A. Prough and D.M. Zeigler. The relative participation of liver microsomal amine oxidase and cytochrome P-450 in N-demethylation reactions. *Arch. Biochem. Biophys.*, 180:363-373, 1977.
- [131] H. Pyysalo and K. Antervo. GC-profiles of chlorinated terpenes (toxaphenes) in some Finnish environmental samples. *Chemosphere*, 14:1723-1728, 1985.
- [132] H. Pyysalo, K. Wickstrom, and R. Litmanen. Contents of chlordane, PCB and DDT compounds and the biotransformation capacity of fishes in the lake area of eastern Finland. *Chemosphere*, 10:865-876, 1981.
- [133] K. Rainio, R.R. Linko, and L. Ruotsila. Polycyclic aromatic hydrocarbons in mussel and fish from the Finnish archipelago sea. *Bull. Environ. Contam. Toxicol.*, 37:337-343, 1986.

- [134] R.A. Rapaport and S.J. Eisenreich. Atmospheric deposition of toxaphene to eastern North America derived from peat accumulation. *Atmos. Environ.*, 20:2367-2379, 1986.
- [135] K.W. Reinert, J.V. Hunter, and T. Sabatino. Dynamic heated headspace analysis of volatile organic compounds present in fish tissue samples. *J. Agric. Food Chem.*, 31:1057-1060, 1983.
- [136] C.P. Rice and M.S. Evans. Toxaphene in the Great Lakes. In J.O. Nriagu and M.S. Simmons, editors, *Toxic Contaminants in the Great Lakes*, pages 163-194. John Wiley and Sons, 1984.
- [137] A. Rogerson, W.Y. Shiu, G.L. Huang, D. Mackay, and J. Berger. Determination and interpretation of hydrocarbon toxicity to ciliate protozoa. *Aquat. Toxicol.*, 3:215-228, 1983.
- [138] D.J. Sabo and J.J. Stegeman. Some metabolic effects of petroleum hydrocarbons in marine fish. In F.J. Vernberg, A. Calabrese, F.P. Thurberg, and W.B. Vernberg, editors, *Physiological Responses of Marine Biota to Pollutants*, pages 279-287. Academic Press, 1977.
- [139] SAS. *SAS System for Linear Models*. SAS Institute, Cary, North Carolina, second edition, 1986.
- [140] P.J. Schmidt. *Analysis of freshwater fishes from Canadian interior provinces*, volume 9 of *Indust. Res. Mem.*, pages 1-5. Fish. Res. Bd. Canada, Pacific Fisheries Experiment Sta., Vancouver, 1948.
- [141] P.J. Schmidt. *Analysis of freshwater fishes from Canadian interior provinces (Cont'd)*, volume 12 of *Indust. Res. Mem.*, pages 1-10. Fish. Res. Bd. Canada, Pacific Fisheries Experiment Sta., Vancouver, 1949.
- [142] P.J. Schmidt. *Analysis of freshwater fishes from Canadian interior provinces*, volume 13 of *Indust. Res. Mem.*, pages 1-7. Fish. Res. Bd. Canada, Pacific Fisheries Experiment Sta., Vancouver, 1950.
- [143] C.J. Schmitt, M.A. Ribik, J.L. Ludke, and T.W. May. National pesticide monitoring program: Organochlorine residues in freshwater fish, 1976-79. Resource Pub. 152, U.S. Dept. Interior, Washington, D.C., 1983.
- [144] B. Schoene, R.A. Fleischman, H. Remmer, and H.F. Olderschausen. Determination of drug metabolising enzymes in needle biopsies of human liver. *Eur. J. Clin. Pharmacol.*, 4:65-73, 1972.
- [145] C.J. Sindermann. Pollution-associated diseases and abnormalities of fish and shellfish: A review. *Fish. Bull.*, 76:717-749, 1979.
- [146] G.P. Slater and V.C. Blok. Isolation and identification of odourous compounds from a lake subject to cyanobacterial blooms. *Wat. Sci. Technol.*, 15:229-240, 1983.
- [147] L.I. Smirnova. Blood indices of the burbot during prolonged total fasting and subsequent feeding. *Akad. Nauk SSSR Doklady, Biol. Sci.*, 160:107-109, 1966.
- [148] J.D. Smith, J. Bagg, and B.M. Bycroft. Polycyclic aromatic hydrocarbons in the clam *Tridacna maxima* from the Great Barrier Reef, Australia. *Environ. Sci. Technol.*, 18:353-358, 1984.

- [149] J.R. St. Amant, M.E. Pariso, and T.B. Sheffy. Polychlorinated biphenyls in seven species of Lake Michigan fish, 1971-1981. In J.O. Nriagu and M.S. Simmons, editors, *Toxic Contaminants in the Great Lakes*, pages 311-320. John Wiley and Sons, New York, 1984.
- [150] J.N. Stein, C.S. Jessop, T.R. Porter, and K.T.J. Chang-Kue. *Fish resources of the Mackenzie river valley*, volume II of *Interim Report. Fisheries Service*. Environmental-Social Committee, Northern Pipelines, Task Force on Northern Oil Development, Winnipeg, 1973. 260 pg.
- [151] W.M.J. Strachan and C.J. Edwards. Organic pollutants in Lake Ontario. In J.O. Nriagu and M.S. Simmons, editors, *Toxic Contaminants in the Great Lakes*, pages 239-264. John Wiley and Sons, New York, 1984.
- [152] J.R. Sullivan and D.E. Armstrong. Toxaphene status in the Great Lakes. Priority Pollutants Status Report No. 2 WIS-SG-85-241, University of Wisconsin Sea Grant Institute, 1985.
- [153] B. Tan and P. Melius. Polynuclear aromatic hydrocarbon metabolism in fishes. *Comp. Biochem. Physiol.*, 83C:217-224, 1986.
- [154] S. Tanabe, H. Tanaka, and R. Tatsukawa. Polychlorobiphenyls, sDDT, and hexachlorochlorohexane isomers in the western north Pacific ecosystem. *Arch. Environ. Contam. Toxicol.*, 13:731-738, 1984.
- [155] C.E. Thurston, M.E. Stansby, N.L. Karrick, D.T. Miyauchi, and W.C. Clegg. Composition of certain species of fresh-water fish. II. Comparative data for 21 species of lake and river fish. *Food Res.*, 24:493-502, 1959.
- [156] W.G. Tidmarsh, R. Ernst, R. Ackman, and T. Farquharson. Tainting of fishery resources. Report 021, Env. Studies Revolving Fund, Ottawa, 1986. 174 pg.
- [157] P.T.P. Tsui and P.J. McCart. Chlorinated hydrocarbon residues and heavy metals in several fish species from the Cold Lake area in Alberta, Canada. *Intern. J. Environ. Anal. Chem.*, 10:277-285, 1981.
- [158] J.F. Uthe and E.G. Bligh. Preliminary survey of heavy metal contamination of Canadian freshwater fish. *J. Fish. Res. Board Canada*, 28:786-788, 1971.
- [159] J. van Cantfort, J. DeGraeve, and J.E. Gielen. Radioactive assay for aryl hydrocarbon hydroxylase. Improved method and biological importance. *Biochem. Biophys. Res. Commun.*, 79:505-512, 1977.
- [160] W.L.F. Van den Broek. The effects of *Lernaeocera branchialis* on the *Merlangius merlangus* population in the Medway estuary. *J. Fish. Biol.*, 13:709-715, 1978.
- [161] U. Varanasi and D.C. Malins. Metabolism of petroleum hydrocarbons: accumulation and biotransformation in marine organisms. In D.C. Malins, editor, *Effects of Petroleum on Arctic and Subarctic Marine Environments and Organisms. Vol. II. Biological Effects*, pages 175-270. Academic Press, New York, 1977.
- [162] G.D. Veith, D.W. Kuehl, F.A. Puglisi, G.E. Glass, and J.G. Eaton. Residues of PCB's and DDT in the western Lake Superior ecosystem. *Arch. Environ. Contam. Toxicol.*, 5:487-499, 1977.

- [163] P.N. Vijan and G.R. Wood. An automated submicrogram determination of selenium in vegetation by quartz tube furnace atomic absorption spectrophotometry. *Talanta*, 23:89-94, 1976.
- [164] M.J. Vodick, C.R. Elcombe, and J.J. Lech. The effects of various types of inducing agents on hepatic microsomal monooxygenase activity in rainbow trout. *Toxicol. Appl. Pharmacol.*, 59:364-374, 1981.
- [165] R. Wagemann. Heavy metals in fish from the Mackenzie River at Fort Good Hope (April, 1985 sampling). Report, 10 pg, 1985.
- [166] D.G. Walton, W.R. Penrose, and J.M. Green. The petroleum-inducible mixed-function oxidase of cunner (*Tautoglabrus adspersus* Walbaum 1792): some characteristics relevant to hydrocarbon monitoring. *J. Fish. Res. Board Can.*, 35:1547-1552, 1977.
- [167] B. Wheatley. Methylmercury in Canada. Exposure of Indian and Inuit residents to methylmercury in the Canadian environment. Report, Health and Welfare Canada, Medical Services Branch, 1978.
- [168] J.A. Whipple, M.B. Eldridge, and P. Benville Jr. An ecological perspective of the effects of monocyclic aromatic hydrocarbons on fishes. In F.J. Vernberg, A. Calabrese, F.P. Thurberg, and W.B. Vernberg, editors, *Biological monitoring of marine pollutants*, pages 483-551. Academic Press, New York, 1981.
- [169] R.D. Wilson, P.H. Monaghan, A. Osanik, L.C. Price, and M.A. Rogers. Estimate of annual input of petroleum to the marine environment from natural seepage. *Trans. Gulf Coast Assoc. Geol. Soc.*, 23:182-193, 1973.
- [170] D.M. Woltering. The growth response in fish chronic and early life stage toxicity tests: a critical review. *Aquat. Toxicol.*, 5:1-21, 1984.
- [171] C.S. Wong, W.J. Cretney, R.W. MacDonald, and P. Christensen. Hydrocarbon levels in the marine environment of the southern Beaufort Sea. Beaufort Sea Project, Tech. Rep. 38, Institute of Ocean Sciences, Sydney, B.C., 1976. 113 pg.
- [172] M.P. Wong. Chemical residues in fish and wildlife species harvested in northern Canada. Northern Affairs Program Environmental Studies No. 46, Northern Environment Directorate, Indian and Northern Affairs Canada, Ottawa, 1985.
- [173] D.F. Woodward, P.M. Mehrle Jr., and W.L. Mauk. Accumulation and sublethal effects of a Wyoming crude oil in cutthroat trout. *Trans. Amer. Fish. Soc.*, 110:437-445, 1981.
- [174] D.F. Woodward, R.G. Riley, and C.E. Smith. Accumulation, sublethal effects, and safe concentration of a refined oil as evaluated with cutthroat trout. *Arch. Environ. Contam. Toxicol.*, 12:455-464, 1983.
- [175] M. Zell and K. Ballschmiter. Baseline studies of the global pollution II. Global occurrence of hexachlorobenzene (HCB) and polychlorocamphenes (toxaphene) (PCC) in biological samples. *Fresenius Z. Anal. Chem.*, 300:387-402, 1980.

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